Dolutegravir in Pregnant HIV Mothers and their Neonates

DolPHIN-2 – Dolutegravir in Pregnant HIV mothers and Neonates

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GCP Compliance Statement

This trial will be conducted in compliance with the protocol, with the principles of ICH Guideline E6 for Good Clinical Practice, the Declaration of Helsinki, and all applicable regulatory requirements.

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ABBREVIATIONS

ABC	Abacavir
ACTG	AIDS Clinical Trials Group
AE	Adverse event
ALT	Alanine transaminase
ARV	Antiretroviral
AST	Aspartate transaminase
ATV	Atazanavir
bd	Twice daily
BM	Breast milk
cART	Combination antiretroviral therapy
CI	Confidence interval
CL	Clearance
CNS	Central Nervous System
CPK	Creatine phosphokinase
CRF	Case report form
CV	Coefficient of variation
CYP	Cytochrome P450
DALY	Disability adjusted life years
DDIs	Drug drug interactions
DILI	Drug Induced Liver Injury
DMPA	Depomedroxyprogesterone
DRV	Darunavir
DTG	Dolutegravir
EFV	Efavirenz (an NNRTI)
ENG	Etonogestrel
EPDS	Edinburgh Postnatal Depression Scale
EVG	Elvitegravir
FDA	U.S. Food and Drug Administration
FM	Foetal-maternal
FTC	Emtricitabine
GCP	Good clinical practice
GFR	Glomerular filtration rate
GI	Gastrointestinal
HADS	Hospital Anxiety and Depression Scale
Hb	Haemoglobin
HBV	Hepatitis B
HCV	Hepatitis C
HELLP	Hypertension Elevated Liver enzymes Low Platelets
HIV	Human immunodeficiency virus
HSR	Hypersensitivity Reaction

IDSMB	Independent Data Monitoring Committee
IGMST	Infant Gross Motor Screening Test
INSTI	Integrase strand transfer inhibitor
IP	Investigational product
IRIS	Immune reconstitution inflammatory syndrome
ITT	Intention to treat
LFTs	Liver function tests
LNG	Levonorgestrel
LNMP	Last normal menstrual period
MTCT	Mother to child transmission
NGO	Non-governmental organisation
NNRTI	Non-nucleoside reverse transcriptase inhibitor
NOAEL	No observed adverse effect level
NRTI	Nucleoside reverse transcriptase inhibitor
NTD	Neural Tube Defect
NTP	National treatment programme
NVP	Nevirapine (an NNRTI)
PD	Pharmacodynamic
PIL	Partner Information Leaflet
PK	Pharmacokinetic
PMTCT	Prevention of mother-to-child transmission of HIV
PRSAE	Possible suicidality related adverse event
Qd	Once-daily
RAL	Raltegravir
RCT	Randomized controlled trial
RTV	Ritonavir
SAE	Serious adverse event
SoC	Standard of care
SSA	Sub-Saharan Africa
SUSAR	Suspected unexpected adverse reaction
TAF-LD.	Tenofovir alafenamide/lamivudine/dolutegravir
TB	Tuberculosis
tCTU	Tropical Clinical Trials Unit
TDF	Tenofovir disoproxil fumarate
TLD	Tenofovir disoproxil fumarate/lamivudine/dolutegravir
TLE	tenofovir disoproxil fumarate/ lamivudine/ efavirenz
TMG	Trial management group
TSC	Trial Steering Committee
UGT	Uridine diphosphate glucuronosyltransferase
ULN	Upper limit of normal
Vd	Volume of distribution
VL	HIV viral load

WHO	World Health Organization
ZDV	Zidovudine

1. CONTENTS

Contents

2. PROTOCOL SYNOPSIS

The original aims of the DolPHIN 2 study, and the updated aims of the DolPHIN-2 TRIO extension are given in this protocol. The original protocol content has not been altered.

Updates to study procedures and activities for the TRIO Extension are given at the end of each section, in blue boxes

Aim

To evaluate dolutegravir (DTG) efficacy in women who present with untreated HIV in late pregnancy

Rationale

In developing countries many women present with a new HIV diagnosis in late pregnancy, and are at high risk of transmitting infection during delivery because existing treatments do not reduce the HIV viral load sufficiently quickly. Moreover, women may acquire non-nucleoside reverse transcriptase inhibitor (NNRTI) resistance from primary transmission, or use of nevirapine (NVP) in previous pregnancies. In these circumstances, DTG is likely to be more effective in reducing mother to child transmission (MTCT) of HIV than NNRTI-based regimens.

Study design

An open-label, multi-centre randomised controlled trial of DTG vs efavirenz-based regimens for women commencing cART in late pregnancy. HIV positive pregnant women presenting with untreated HIV infection in late (≥28 weeks gestation) pregnancy will be randomised 1:1 to receive DTG (50mg once daily) + 2 nucleoside reverse transcriptase inhibitors (NRTIs) or EFV + 2 NRTIs (SoC)

Inclusions

- Evidence of informed consent
- Willing to participate,
- Women aged 18 years and above
- Pregnant (≥28 weeks gestation by best available gestation estimation)
- Untreated HIV infection in late pregnancy (diagnosed using nationally recommended rapid tests)

Exclusions

- Received any antiretroviral drugs in previous 12 months
- Ever received integrase inhibitors
- Previous documented failure of an NNRTI-containing ART regimen, previous EFV-associated toxicity or other clinical history that would preclude randomisation based on investigator judgement
- HIV viral load < 50 copies/ml at enrolment (pre-ART)
- Serum hemoglobin <8.0 g/dl
- eGFR<50 ml/min

- Elevations in serum levels of alanine aminotransferase (ALT) >5 times the upper limit of normal (ULN) or ALT >3xULN and bilirubin >2xULN (with >35% direct bilirubin).
- History or clinical suspicion of unstable liver disease (as defined by the presence of ascites, encephalopathy, coagulopathy, hyperbilirubinaemia, oesophageal or gastric varices or persistent jaundice).
- Participant with severe pre-eclampsia (e.g. HELLP (Hypertension Elevated Liver enzymes Low Platelets)), or other pregnancy related events such as renal or liver abnormalities (e.g. grade 2 or above proteinuria, total bilirubin, ALT or AST) at the time of enrolment
- Paternal objection for infant participation in DTG arm (where disclosure has taken place – see below) – applies to Uganda site only
- Medical, psychiatric or obstetric condition that might affect participation in the study based on investigator judgement
- Receiving any of the following medications (current or within past 2 weeks): anti-epileptic drugs, TB therapy or other drugs known to significantly interact with EFV or DTG

Primary Endpoint

- Occurrence of maternal HIV viral load <50 copies at delivery

Secondary Endpoints & Exploratory endpoints related to Efficacy

- Plasma VL <1000 copies at delivery
- VL dynamics in breast milk and maternal plasma
- Maternal VL response to 48w (% under 50 and 1000 copies)
- Occurrence of MTCT up 72w
- DTG exposure in maternal plasma, breast milk and infants
- Virological resistance to cART
- Viral dynamics and pharmacokinetics in the genital tract of mothers, and screen for co-infections by examining the vaginal microbiota
- Cost-effectiveness of DTG in South African and Ugandan pregnant women

Safety Endpoints

Primary safety endpoint:

- Drug toxicities defined according to DAIDS criteria

Other safety endpoints:

- Safety and tolerability of DTG in mother
- Safety of DTG in infant (death or injury, congenital anomalies, weight, anthropometric measurements for gestational age, pre-term delivery, growth trajectories, infant gross motor development)

Number recruited

N=125 per arm; total 250 mothers

Study Sites

1) Infectious Diseases Institute, Makerere University College of Health Sciences, Kampala, Uganda

2) Gugulethu Community Health Centre, Cape Town, South Africa

2.1 TRIO EXTENSION PROTOCOL SYNOPSIS

Aim	To evaluate the long-term risk-benefit for participants who were randomised to dolutegravir compared with efavirenz, as an extension to the DolPHIN 2 study.
Rationale	In the DolPHIN 2 study poorer maternal clinical outcomes on DTG-containing regimens were identified. Despite faster initial HIV-RNA suppression on DTG, there have been more cases of vertical HIV transmission, infant deaths and stillbirths in the DTG arm compared with EFV. Adverse birth outcomes for DTG-treated mothers might be linked with clinical obesity – this needs to be investigated further.
Study design	DolPHIN-2 TRIO is an open-label, multi-centre trial of dolutegravir versus efavirenz-based regimens for women randomised in the third trimester of pregnancy, and continuing combination antiretroviral therapy after pregnancy.
Inclusions	• Evidence of informed consent • Willing to participate, • Women aged 18 years and above • Previous randomisation to the DolPHIN 2 study
Exclusions	• Medical, psychiatric or obstetric condition that might affect participation in the study based on investigator judgement
Primary Endpoint	HIV Viral load at 192 weeks <50 copies per mL
Secondary Endpoints & Exploratory endpoints related to Efficacy	Efficacy of Treatment (HIV viral load < 1000 copies per mLat 192 weeks) Adherence to treatment regimen Weight gain • associated risk for maternal health • Occurrence of Mother to Child transmission at first TRIO visit
Safety Endpoints	Primary safety endpoint: • Drug toxicities defined according to DAIDS criteria (maternal & infant) **Key clinical events:** • Diabetes (as defined by HbA1c > 6.5%, Fasting glucose >7.0 mmol/L (126 mg/dL), Random glucose > 11.1 mmol/L (200 mg/dL)

	• Myocardial Infarction • Birth Defects (unreported in index pregnancies, and subsequent pregnancies) • HIV MTCT child transmission (at first trio visit and in subsequent pregnancies) • Radiologically confirmed fractures Other safety endpoints: • Laboratory end points (eg, lipids, glucose) • Blood Pressure changes
Number recruited	Recruitment is open to the recruited patients from the DolPHIN 2 study (268) including those classed as 'withdrawn' or 'lost to follow up'. Participants that withdrew consent will not be considered.
Study Sites	1) Infectious Diseases Institute, Makerere University College of Health Sciences, Kampala, Uganda 2) Gugulethu Community Health Centre, Cape Town, South Africa

3. LAY SUMMARY

Despite global prevention initiatives, mother-to-child transmission (MTCT) remains a major route of HIV infection. In sub-Saharan Africa (SSA) over 1 in 5 HIV-infected pregnant women are diagnosed in late pregnancy each year, and represent the group most likely to transmit HIV infection to their infants due to delayed initiation of combination antiretroviral therapy (cART). Since risk of MTCT is directly related to maternal HIV viral load (VL), antiretroviral drugs (ARVs) which result in very fast rates of VL decline may be more effective at preventing MTCT (PMTCT) of HIV, and make the global goal of elimination of MTCT more likely to be achieved.

Dolutegravir (DTG) is a new ARV which results in significantly faster VL declines when compared to efavirenz (EFV)-based regimens (the standard-of-care for SSA). The median time to achieving undetectable VL in non-pregnant adults was 28 versus 84 days, with differences most apparent in the first 12 weeks of therapy. DTG was shown to be superior to EFV with improved efficacy and tolerability through 48 weeks, and this drug is expected to retain activity in the presence of archived resistance associated with prior short course or single dose antiretroviral drugs in previous pregnancies. Finally, DTG can be scaled up for generic production at a cost comparable with the cheapest ARVs. As a result DTG is expected to become widely available throughout SSA. However, data on efficacy and safety in pregnancy are lacking.

We propose an open-label, randomised controlled trial of DTG versus EFV -based regimens for 250 women commencing cART in late pregnancy, randomised 1:1 to DTG vs EFV-based cART. The purpose of this study is to inform treatment guidelines and for the first time specifically address the treatment needs of this group of women- hence the trial is powered for superiority over EFV. The primary endpoint is maternal VL at delivery, with secondary endpoints including

safety and tolerability of DTG in both mother and infant, VL decline in breast milk, development of drug resistance, pharmacokinetics of DTG in mother-infant pairs, pharmacogenomics factors relating to efficacy or toxicity of DTG, and MTCT of HIV up to 72 weeks postpartum. Two sites have been selected – Infectious Diseases Institute, Makerere University, Kampala, Uganda and the University of Cape Town, South Africa – both have a strong track record of successfully delivering collaborative multidisciplinary research in PMTCT. Furthermore, we will conduct health economics analysis to examine costs and cost-effectiveness of DTG in late-presenting pregnant women.

The desired outcome of this project is to establish high quality evidence and operational guidance for use of DTG in late pregnancy. Late-presenting HIV-infected pregnant women are an important, but neglected group of vulnerable individuals in whom a randomised controlled intervention of HIV treatment has never previously been undertaken. We will work closely with WHO and the Clinton Health Access Initiative to ensure successful delivery of the project objectives.

3.1 TRIO EXTENSION LAY SUMMARY

The DolPHIN 2 trial has met its exploratory objectives of increasing the evidence base for optimal ARVs in first-line treatment for HIV, and this has been shared with the World Health Organization to support updates to the HIV Treatment Guidelines. However, over the course of the last two years, emergent safety issues with dolutegravir (DTG) containing first-line regimens have been observed, necessitating a continuation of trial monitoring activities beyond the 96 weeks initially planned for the trial.

These safety signals related to the use of DTG with or without tenofovir alafenamide (TAF) were identified in 2019. In the ADVANCE (Venter WDF, Moorhouse M, Sokhela S, et al.) and NAMSAL (NAMSAL ANRS 12313 Study Group, Kouanfack C, Mpoudi-Etame M, et al.) trials people taking DTG had an elevated risk of developing clinical obesity (presented at the IAS Conference in July 2019, and reported in the *New England Journal of Medicine* in July 2019). The mean body weight at 48 weeks continued to rise in women taking DTG, with no sign of a plateau. In the DolPHIN-2 trial, there were more cases of vertical HIV transmission and adverse birth outcomes for pregnant women taking DTG, versus those taking efavirenz (EFV).

Clinical obesity increases the risk of myocardial infarction, diabetes and adverse birth outcomes. Reports on increased metabolic conditions in DTG-early adopter countries are also emerging. In Uganda, there have been reports of hyperglycaemia and treatment-emergent diabetes for people switching from EFV to DTG-based regimens. As plans for scale-up of DTG are accelerating, there is the need to expand the knowledge base of these key safety issues, which remain unresolved and could worsen over time if body weight continues to rise.

People with HIV are treated with ARVs for life. It is therefore important to ensure that any safety signals seen on new ART are followed up until they either resolve or stabilise. If we

can follow up the participants for a total of 4 years of overall randomised treatment (since the projects started), the assessment of the overall effects of clinical obesity on a range of associated diseases would be enabled.

4. ETHICAL JUSTIFICATION, AND RISK-BENEFIT ASSESSMENT

4.1 Ethical Justification

Universal HIV testing of pregnant women in low/middle income countries continues to detect new HIV diagnoses, and many of these mothers present late in pregnancy (≥28 weeks gestation). Maternal VL at delivery is a key predictor of the risk of MTCT (Garcia, Kalish et al. 1999), and existing EFV-based regimens are unable to reduce the viral load sufficiently rapidly among late presenting mothers despite widespread rollout of WHO Option B+ (WHO 2010, WHO 2012, WHO 2013).

The justification for studying DTG in pregnancy includes:

a) **Rapid viral load drop potentially beneficial in late diagnosis during pregnancy**. In SSA, pregnant mothers tend to engage with health services later in pregnancy compared with Europe, and new HIV diagnoses resulting from universal testing at ≥28w gestation are frequent (Myer, Phillips et al. 2015). DTG results in a very rapid reduction in VL; median time to undetectable VL in the SINGLE study (non-pregnant adults) was 28 days vs. 84 days with Atripla (P<0.0001) (Walmsley, Antela et al. 2012). Use of DTG may lower risk of MTCT of HIV in late presenters (de Ruiter, Taylor *et al*. 2014).

b) **Good tolerability and l**ow **risk for serious drug-drug interactions.** The potential for drug-drug interactions is significantly less for DTG compared with many other ARVs (Patel, Abdelsayed *et al*. 2011) including with hormonal contraception and TB therapy (Dooley, Sayre *et al*. 2013).

c) **Likely widespread availability of generic DTG** in the coming years. Generic manufacture of DTG will make this drug affordable as an alternative first line, or else second line agent (Hill 2013). Clinical guidelines from the US and Europe currently rank integrase strand transfer inhibitors (INSTIs) such as raltegravir (RAL) and elvitegravir (EVG) alongside NNRTIs as preferred first line agents.

d) **Proven efficacy in participant with established drug resistance** to other ARVs. In a cross-sectional study conducted among HIV-1 antiretroviral naïve participant in five African countries, the highest prevalence of transmitted resistance was observed in Kampala, Uganda 12.3% (22 of 179; 7.5-17.1) (Hamers, Wallis *et al.* 2011), with figures of 3% reported for South Africa (Manasa, Katzenstein *et al*. 2012). Studies across Africa have shown that treatment response to NNRTIs in mothers and children exposed to NVP (particularly single dose) during pregnancy is blunted (Musiime, Ssali *et al*. 2009). The SAILING (Cahn, Pozniak *et al.* 2013) and VIKING (Castagna, Maggiolo *et al.* 2014) studies have established the efficacy of DTG in participant with prior drug resistance.

e) **Necessity of investigating safety and efficacy in target population.** Previous experience suggests it is universally the case that pregnant women in SSA receive new ARVs ahead of any proper evaluation, and clinical studies (if undertaken at all) occur at a much later stage

after many mothers and infants have been exposed. Safety of DTG in this population is not yet established (Ford, Mofenson *et al*. 2010), (Sibiude, Madelbrot *et al*. 2013) (Westreich, Rubel *et al*. 2010).

f) **Importance of undertaking the study in SSA.** Any pivotal study if undertaken in developed countries would have limited generalizability to an African setting because i) ARVs tend to be initiated later in gestation in newly presenting pregnant women, ii) the standard of care across SSA is likely to be an EFV or NVP-based regimen, iii) infant breastfeeding is strongly recommended iv) host factors such as BMI, pharmacogenomics, ethnicity etc may impact on pharmacokinetics (PK) of ARVs.

g) **Continued administration of DTG post-partum (whilst breastfeeding**). Under standard of care, the infant will receive 6 weeks of ARV prophylaxis (as NVP with or without zidovudine [ZDV]), and would be exposed to the NNRTI/NRTI/NRTI combination received by the mother under standard of care. In this study, infants will be exposed to DTG via the breastmilk following birth. A recent case study has demonstrated transfer of low concentrations of DTG from mother to infant through breastfeeding (Kobbe, Schalkwijk et al. 2016). We believe this risk to be acceptable given the potential benefits, but data do not yet exist regarding safety of DTG in newborns. Furthermore, the risk is unlikely to exceed any potential risk of in utero exposure to DTG. Drug-drug interactions between directly administered NVP and DTG ingested via the breastmilk are unlikely to have clinical significance (this is being studied in detail in the ongoing DolPHIN-1 study). The potential risks of breastfeeding are fewer than those of giving formula feeding. Replacement of breast feeding with formula feeding would be unethical due to the loss of the immune benefits of colostrum to the neonate, risks intrinsic to formula feeding in low resource settings and increased risk of HIV transmission if mixed feeding occurs (Teasdale et al, 2011) The potential risks of low-level ARV causing drug-resistance in the infant should the PMTCT fail exist for the standard of care NNRTIs and NRTIs; it is not anticipated that DTG will carry a greater risk but nevertheless this is best monitored within a clinical trial setting before widespread uptake without monitoring.

4.2 Risk Assessment for Dolutegravir in Pregnancy

DTG is currently classified as FDA Category B. Pregnant women were excluded from the Phase IIb/III DTG clinical studies and participants who became pregnant were required to discontinue from the studies. Of the 52 pregnancies reported across the DTG clinical studies and compassionate use program through 16 Jan 2016 (ViiV, unpublished data), 23 resulted in delivery of a normal healthy baby (including the partner pregnancy and a twin pregnancy), 13 in elective termination, one pregnancy was ectopic (EFV/tenofovir [TDF]/ emtricitabine [FTC]) and 7 resulted in spontaneous abortions (3 for DTG, 2 for EFV/TDF/FTC and 1 each for RAL and darunavir (DRV)+ ritonavir (RTV); all between 2 and 10 weeks gestation). In seven cases, the pregnancy was either ongoing or unknown (all DTG). During one pregnancy, involving an ART-experienced (INSTI-naïve) participant exposed to DTG plus abacavir (ABC), atazanavir (ATV) and RTV, a routine ultra sound at 31 weeks gestation indicated a congenital anomaly of double outlet right ventricle with ventricular septal defect in the fetus. The events were considered unrelated to investigational product (IP) by the reporting investigator. None of the cases resulting in adverse pregnancy outcome were considered reasonably attributable to IP by

the reporting investigators.

Neural tube defect (NTD) in infants born to women with exposure to DTG at the time of conception has been identified from a preliminary unscheduled analysis in the Tsepamo study, Botswana (4 NTD cases out of 426 pregnancies). The overall incident rate for NTDs with DTG exposure is 0.9 % compared to a background incidence of 0.1 %. Data from other sources that include the antiretroviral pregnancy register, clinical trials and postmarketing use have not indicated a similar potential safety issue. There are currently no congenital abnormality signals associated with the use of DTG during pregnancy from other data sources.

The antiretroviral pregnancy registry (http://www.apregistry.com/forms/interim_report.pdf) provides data on 22 first trimester and 29 second and third trimester exposures with two anomalies detected (data up to Jan 2016). There have been no NTDs with DTG exposure reported to the antiretroviral pregnancy registry (May 2018).

The overall nonclinical reproductive and developmental toxicity profile for DTG in rats and rabbits suggests that DTG is not teratogenic and has a low potential for fetal risk. There were no effects on fertility or early embryonic development in rats orally administered DTG at ~1000 mg/kg/day in males or females. The no adverse effect level (NOAEL) was 1000 mg/kg/day, which corresponds to ~33X the expected human exposure for a 50 mg once daily, based on gender averaged mean exposures achieved in the 4 week rat toxicity study. No adverse effects on fetal development were observed in pregnant rats orally administered DTG at ~1000 mg/kg/day. The NOAEL for maternal and fetal toxicity was 1000 mg/kg/day, which corresponds to ~38X the expected human exposure for a 50 mg once daily dose. In an embryofetal development (EFD) study in rabbits, DTG was orally administered at 40, 200, or 1000 mg/kg/day to pregnant rabbits. Suppressed body weight gain (13.6% on gestation Day 19), decreased food consumption (up to 53%) and scant or no faeces/urine associated with the decreased food consumption were noted in the 1000 mg/kg/day dams. Maternal toxicity at this dose precluded dosing DTG at higher doses in rabbits because maternal toxicity can confound teratogenicity assessment. Therefore, the dose of 1000 mg/kg/day was the maximal dose that could be administered in this embryo fetal development EFD study.

The NOAEL was 200 mg/kg/day for maternal general toxicity (~0.27X the expected human exposure for a 50 mg once daily dose) and 1000 mg/kg/day for maternal reproductive function and embryofetal development (0.56X the expected human exposure for a 50 mg once daily dose,). Of note, there were no teratogenic effects at 1000 mg/kg/day, a dose that exceeded the NOAEL for maternal toxicity. In a pre-and postnatal development study, DTG was administered to female rats at doses of 5, 50 or 1000 mg/kg/day from Day 6 of gestation to Day 20 of lactation. Suppressed body weight gain and decreased food consumption were noted in dams (F0) in the 1000 mg/kg/day group during the lactation period, which were associated with mild decreases in body weights in the offspring in the 1000 mg/kg/day group from pre-weaning until adolescence. There were no adverse effects on maternal pregnancy, parturition, lactation or offspring (F1) survival, behavioral or reproductive function. The NOAEL for maternal reproductive function was 1000 mg/kg/day (~32X above the expected human exposure for a 50 mg once daily dose,

based on exposures achieved in female rats in the 4 week toxicity study). Due to the decreased body weights of the offspring observed at higher doses, the NOAEL for pre- and postnatal development of the offspring (F1) was 50 mg/kg/day. This is ~25X above the expected human exposure for a 50 mg once daily dose (extrapolated from gender mean exposures achieved in the rat 14 day toxicity study). Based on the fact that effects on offspring body weights were noted at doses where maternal toxicity was observed, and the presence of considerable safety margins expected at the proposed clinical doses, there is minimal risk for adverse effects on postnatal development in offspring of mothers receiving DTG.

DTG is excreted in the milk of lactating rats. Following oral administration (50 mg/kg) to lactating rats on Day 10 postpartum, total radiocarbon concentrations in milk were up to 2-fold greater than those in maternal blood. The metabolite profile of milk indicated that parent DTG represented more than 95% of the total radiocarbon, consistent with the findings in plasma from female rats in an earlier study. These data suggest that F1 offspring in the pre- and postnatal toxicity study were exposed to the drug via the milk. Following oral administration of DTG (50 mg/kg) to pregnant rats on Day 18 post conception, DTG-related material was found, by quantitative whole body autoradiography analysis, to be widely distributed to the fetuses over the 24- hour sampling period. These data indicate that DTG is able to cross the placental barrier.

DTG is primarily metabolized through UGT1A1 with minimal renal excretion (<1% of total dose given orally). DTG is a drug with low clearance (~1 L/hr for CL/F after oral dosing), low volume of distribution (~12.5 L for Vd/F after oral dosing), and high plasma protein binding (>99%). Physiological changes during pregnancy which may impact on DTG PK (Table 1) are being evaluated in the DolPHIN-1 study.

Table 1: Potential pharmacokinetic changes during pregnancy

Possible Physiological Changes During Pregnancy*	**Potential Impact on DTG PK**
Changes in total body weight and body fat composition.	Increase clearance (CL) as well as the volume of distribution (Vd)
Delayed gastric emptying and prolonged gastrointestinal transit time.	Increase oral bioavailability, delayed time to observed maximal drug concentration (tmax)
Increase in extra cellular fluid and total body water.	Increase Vd
Increased cardiac output, increased stroke volume, and elevated maternal heart rate.	Unlikely to have effect
Decreased albumin concentration with reduced protein binding.	Increase unbound fraction, CL, and Vd
Increased blood flow to the various organs (e.g., kidneys, uterus).	Unlikely to have effect
Increased glomerular filtration rate.	Unlikely to have effect
Changed hepatic enzyme activity, including phase I CYP450 metabolic pathways (e.g., increased CYP2D6 activity), xanthine oxidase, and phase II	May affect CL

metabolic pathways (e.g., N-acetyltransferase	

*FDA Guidance for Industry (2004): Pharmacokinetics in Pregnancy — Study Design, Data Analysis, and Impact on Dosing and Labeling.

DTG has shown to be not prone to clinically significant changes by demographic factors (age, gender, weight, etc) based on population PK analyses (Zhang, Hayes et al. 2015). Accumulated safety data showed that DTG is in general well tolerated and no PK/Pharmacodynamic (PD) relationship for safety measures has been identified. Therefore, despite the potential impacts of physiological changes on DTG PK, it is expected that DTG PK should not change significantly during pregnancy. The clinical dose of 50mg once daily will be used in this study.

4.3 Role of DTG in National Policy (Uganda and South Africa)

Dolutegravir (as TDF-3TC + DTG) has been included in the Consolidated Guidelines for Prevention and Treatment of HIV in Uganda (December 2016) as an alternative first-line regimen for adults and adolescents who have a condition where EFV is contra-indicated, including severe clinical depression, psychosis or suicidality; ongoing complications of neurological disease which make it impossible to assess for side-effects of EFV; use of anxiolytics especially benzodiazepines or carbamazepine; severe hepatic impairment; intercurrent TB treated with bedaqualine and situations where the only available family planning is interacting hormonal methods such as levonorgestrel, ethinyl oestradiol or etonorgestrel.

In South Africa, DTG is licensed and available in the private sector and is rapidly gaining recognition as a preferred agent. It is listed as a possible first line ART agent in the 2015 revision of the Southern African HIV Clinicians Society Guidelines (Meintjes, Black *et al.* 2015), and is listed as a preferred first line agent in the 2017 revision of these guidelines. Currently, neighbouring Botswana is currently using DTG as the preferred first-line agent in public sector treatment programmes, and it is anticipated that South African National Department of Health guidelines, currently under revision, will include DTG as a first-line agent with public sector availability during 2018.

4.4 Risk Management Plan

Table 2 sets out ethical considerations and safeguards which will be used in a series of community, clinical and scientific consultations:

Table 2 Risk Management Plan

Potential Risk	**Details**	**Risk Mitigation Strategy**
Risk of harm to fetus from teratogenic effects of drug	With other ART agents, there are concerns with regard to teratogenicity (e.g. EFV) and maternal hepatic/rash toxicity (i.e. NVP). ZDV, ABC and 3TC have high level of transplacental transfer. Data available for teratogenic risk for DTG is described in Section 4.2. Preliminary data from the Tsepamo study in Botswana have reported an incidence of 0.9 % of neural tube defects (NTDs) amongst infants born to women receiving DTG at the time of becoming pregnant or early on in the first trimester. No NTDs have been reported in women starting DTG after the first trimester.	Enrolling women after week 28 of gestation avoids exposure during the period of organogenesis. There is no specific mitigation of risk to the fetus. However, the risk is considered low based on available data for DTG. Frequent assessment of clinical and laboratory parameters (via this protocol), and routine care by appropriate provider of healthcare to pregnant women. Contraception will be offered to all women post-partum and confirmed at each study visit. Women who decline contraception and are receiving DTG will be switched to EFV based ART.
Pregnancy can increase risks of HIV progression, complications and perinatal transmission	The safe use of DTG in human pregnancy has not been established. DTG has been shown to cross the placenta in reproductive toxicity studies	Mitigated by ART in both experimental and control arms. Additionally women taking ART during pregnancy can reduce risk of transmission to infant (i.e. early control of viral replication) (Meyers, Qian *et al.* 2015)
Hypersensitivity reactions (HSR) and rash	HSR is uncommon with DTG. Rash was reported in DTG Phase IIb/III clinical trials; episodes were	Specific/detailed toxicity management guidance is provided for suspected HSR with DTG (see

	generally mild to moderate in intensity; no episodes of severe rash, Stevens-Johnson Syndrome, TEN and erythema multiforme were reported.	Section 11.3.2) or rash with DTG (see Section 11.3.3)
Drug induced liver injury (DILI) and other clinically significant liver chemistry elevations	Non-clinical data suggested a possible low risk for hepatobiliary toxicity with DTG. Drug-related hepatitis is uncommon with DTG regardless of dose or treatment population. For participants with hepatitis B (HBV) virus and/or hepatitis C (HCV) virus co-infection, immune recovery, and HBV coinfection likely contributed to elevations in liver chemistries.	Participants meeting the following criteria during the screening period are excluded from participating (Section 6.3). Liver-stopping criteria and management are specified a priori See Section 11.3.1)
Gastrointestinal (GI) intolerance	Mild to moderate GI intolerance (mainly diarrhoea and nausea) is associated with DTG; however there were no indications of an increased risk for peptic ulcers or serious erosions.	Routine monitoring of GI symptoms will be performed.
Renal function	Mild elevations of creatinine with DTG are related to a benign effect on creatinine secretion. DTG has been shown to have no significant effect on glomerular filtration rate (GFR) or effective renal plasma flow. DTG is eliminated by renal excretion and exposure increases in participant with renal dysfunction.	Participants with a Calculated eGFR <50ml/min will have modification of NRTI dose as per National Guidelines Specific/detailed toxicity management guidance is provided for participants who develop a decline in renal function and/or proteinuria.
Creatine phosphokinase (CPK) elevations	Asymptomatic CPK elevations mainly in association with exercise have been reported with DTG therapy.	Safety bloods and specific toxicity management guidance is provided (see below)
Immune reconstitution inflammatory syndrome (IRIS)	Rapid HIV-1 RNA decline and early recovery of CD4+ cell may give rise to IRIS.	Participants will have routine laboratory monitoring, and all study physicians are experienced in managing IRIS.
Psychiatric disorders	Psychiatric disorders including suicidal ideation and behaviours are common in HIV infected participant.	Participants who in the investigator's judgment, pose a significant suicidality risk, will be

	The psychiatric profile for DTG (incl. suicidality, depression, bipolar and hypomania, anxiety and abnormal dreams) was similar or favourable compared with other ART. The reporting rate for insomnia was statistically higher for blinded DTG+ABC/3TC compared to EFV/TDF/FTC in ING114467; however, this was not duplicated in any other Phase IIb/III study conducted with DTG.	excluded. Because of the elevated risk in the HIV- infected population, treatment emergent assessment of suicidality will be monitored during this study. Questionnaires will be administered at baseline at intervals thereafter to assess mental state. We have established clear channels of referral for participants who experience clinical depression or signs of suicidal ideation or behaviour.
Hyperglycaemia/diabetes	DTG may induce hyperglycaemia/diabetes. A published case report and clinical trials (SPRING-2, SAILING, SINGLE and VIKING-3) have shown glucose abnormalities.	Participants will be monitored for hyperglycaemia and diabetes through laboratory monitoring.

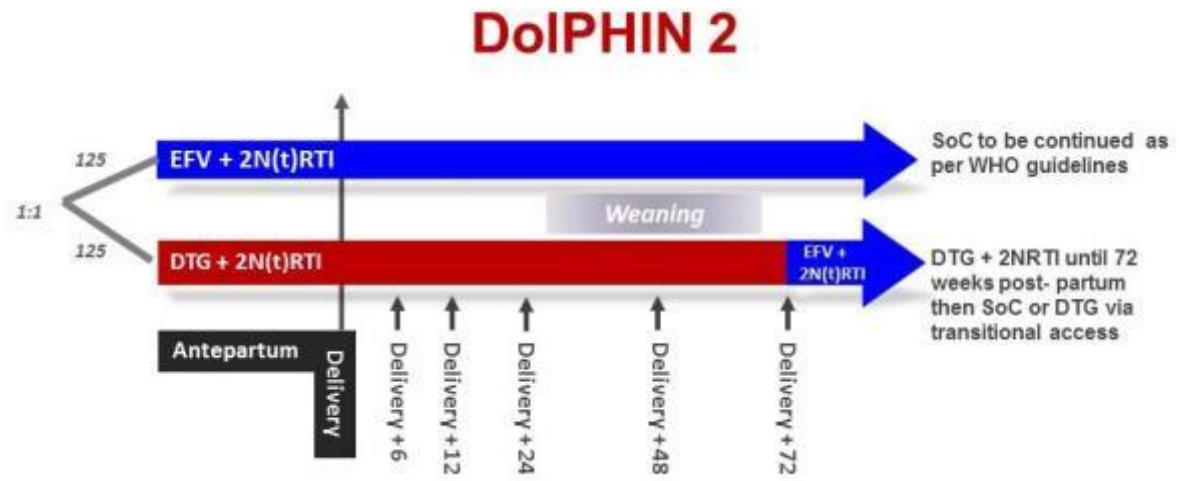
4.5 TRIO EXTENSION ETHICAL JUSTIFICATION & RISK BENEFIT

The principal investigators, IDSMB and community advisory boards have recommended continuing the various arms unchanged, having evaluated the ethics of continuing combinations of similar potency but with complex, evolving and different toxicity.

5. DESIGN, OBJECTIVES AND ENDPOINTS

5.1 Study Design

Open-label, multi-centre randomized controlled trial of DTG in late pregnancy. 250 HIV+ pregnant women (untreated at ≥28weeks gestation) will be randomized 1:1 to receive a DTG-based regimen compared with EFV-based standard of care (regimen not containing INSTI).



5.2 Primary Endpoint

- HIV VL <50 copies/ mL at delivery (+/- 14 days)

5.3 Secondary Endpoints related to Efficacy

- Plasma VL <1000 copies at delivery (+/- 14 days)
- VL dynamics in breast milk and maternal plasma
- Maternal VL response to 48w (% under 50 and 1000 copies)
- Maternal VL response to 72w (% under 50 and 1000 copies)
- Maternal VL (copies/mL) at different visits – repeated measurements (binary and continuous)
- Time from randomisation to the first occurrence of maternal VL (copies/mL) < 50 copies/ mL
- Occurrence of MTCT up 48w
- Occurrence of MTCT at 72w

5.4 Exploratory Endpoints

- DTG exposure in maternal plasma, breast milk and infants
- Virological resistance to cART
- Viral dynamics and pharmacokinetics in the genital tract of mothers, and screen for co-infections by examining the vaginal microbiota

5.5 Safety Endpoints

Primary safety endpoint:

- Drug toxicities defined according to DAIDS criteria

Other safety endpoints:

- Safety and tolerability of DTG in mother
- Safety of DTG in infant (death or injury, congenital anomalies**, weight, anthropometric measurements for gestational age, pre-term delivery, growth trajectories, infant gross motor development)

** structured assessment

5.6 TRIO EXTENSION DESIGN, OBJECTIVES & ENDPOINTS

The TRIO extension to the DolPHIN 2 study will monitor the original randomised participants of the study to follow up to 192 weeks, with 6 monthly visits up to 192 weeks.

1.1. Primary Endpoints

HIV Viral load < 50 copies/mL at 192 weeks

1.2. Secondary Endpoints

Efficacy of Treatment (HIV viral load < 1000 copies at 192 weeks)
Adherence to treatment regimen
Weight gain

- associated risk for maternal health
- occurrence of Mother to Child transmission (measured at first TRIO visit)

1.3. Safety Endpoints

Primary safety endpoint:

- Drug toxicities defined according to DAIDS criteria

Key clinical events:

- Diabetes
- Myocardial Infarction
- Birth Defects (unreported in index pregnancies, and subsequent pregnancies)
- Radiologically confirmed fractures.
- Blood pressure changes

Other safety endpoints:
Changes in lipids, glucose, LFTs,

6. PARTICIPANT POPULATION

6.1 Number of participants

Our sample size calculations are based on clinical trials simulation (SAS v 9.3) using weighted probabilities for achieving VL<50 copies for DTG and EFV in treatment-naïve, non-pregnant adults (Walmsley, Antela *et al*. 2013), Figure 1. Since the period of cART prior to delivery may vary from a single dose to 12w, we have estimated statistical power from simulating five

distributions of gestational age at starting cART (28w-term): i) Normal distribution ii) moderate skew to late gestation (40% ≥38w) iii) moderate skew to early gestation (40% ≤30w) iv) marked skew to late gestation (50% ≥38w) v) marked skew to early gestation (50% ≤30w). Data from Gugulethu (2013-14) suggest that in late presenters iii) is the most likely scenario (Myer, Phillips *et al.* 2015). Allowing for 20% dropout rate, recruitment of 250 HIV-positive women would retain ≥99% power to detect a superiority difference of 28–38% across the five scenarios above of DTG over EFV at the 5% level of significance. For example, for the worst scenario v), with the weighted rate of primary endpoint is 42.0% in the DTG group and 14.1% in the EFV group, the study will have >99% power to detect the absolute difference of 27.9% at significance level of 5% taking into account of 20% drop out rate.

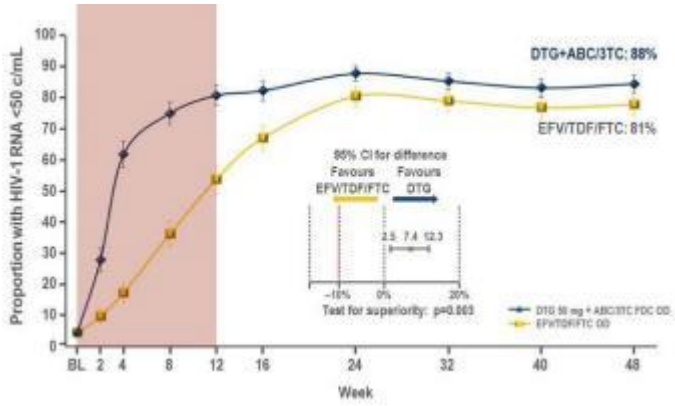


Figure 1: Proportion of participants with viral load <50 copies per mL in the SINGLE study. The shaded area indicates the period of maximum expected benefit in late-presenting pregnant women

An overall sample size of 250 also provides 91% power to detect a 23% difference in the primary safety endpoint (drug toxicity) from 66% in the EFV arm to 43% *(*Walmsley, Antela *et al.* 2013) in the DTG arm (α=0.05, dropout rate=20%) (or 82% to detect a 20% difference from 66% in the EFV arm). Competitive recruitment will take place between the study sites in Uganda and South Africa.

6.2 Inclusion criteria

Participants must meet all of the following inclusion criteria to be eligible for enrolment into the study:

1. Evidence of a personally signed and dated informed consent document indicating that the participant has been informed of all pertinent aspects of the study.
2. Willing and able to comply with scheduled visits, treatment plan, laboratory tests, and other study procedures.
3. Women aged 18 years or older
4. Pregnant (≥28 weeks gestation by best available gestation estimation)
5. Untreated HIV infection in late pregnancy

6.3 Exclusion criteria

Participants presenting with any of the following will not be included in the study:

1) Received any antiretroviral drugs in previous 12 months
2) Ever received integrase inhibitors

3) Previous documented failure of an NNRTI-containing ART regimen, previous EFV-associated toxicity or other history of ARV use that would preclude randomisation based on investigator judgement
4) HIV viral load < 50 copies/ml at enrolment (pre-ART)
5) Serum haemoglobin <8.0 g/dl
6) eGFR<50 ml/min*
7) Elevations in serum levels of alanine aminotransferase (ALT) >5 times the upper limit of normal (ULN) or ALT >3xULN and bilirubin >2xULN (with >35% direct bilirubin).
8) History or clinical suspicion of unstable liver disease (as defined by the presence of ascites, encephalopathy, coagulopathy, hyperbilirubinaemia, oesophageal or gastric varices or persistent jaundice).
9) Severe pre-eclampsia (e.g. HELLP), or other pregnancy related events such as renal or liver abnormalities (e.g. grade 2 or above proteinuria,, total bilirubin, ALT or AST)* at the time of enrolment
10) Paternal objection for infant participation in DTG arm (where disclosure has taken – applies to Uganda site only
11) Medical, psychiatric or obstetric condition that might affect participation in the study based on investigator judgement
12) Receiving any of the following medications (current or within past 2 weeks): anti-epileptic drugs, TB therapy, or other drugs known to significantly interact with either DTG or EFV

*Hb, eGFR, ALT and bilirubin will be undertaken at Screening, with the results made available at the earliest opportunity.

6.3.1 Considerations of Paternal Consent

(Ugandan Site Only with reference to Uganda National Council of Science and Technology [UNCST] guidance)

We recognise that consent of both parents is desirable, however in many cases mothers may choose not to disclose their HIV status to their partner, and consultation with regulatory bodies and community groups have indicated differences in ethical position over the role of paternal consent. In accordance with published guidance from the Ugandan National Council of Science and Technology, the mother shall be asked whether she gives consent for her partner to receive an abbreviated version of the PIL; this shall contain the fundamental details of the study, the contact details of members of the study team and space for a signature to confirm that the father has read and understood this information (this is not consent to participate). Where discrepancies in willingness to participate between the mother and father are recorded, mothers will not be recruited.

In South Africa, only maternal consent is sought, with issues around disclosure, partner testing and counselling managed as per national treatment programme. For both sites, counselling services are provided to all study participants by a dedicated trained counsellor provided by DolPHIN-2.

6.4 Selection and Inclusion of Participants

Participants will be identified and screened at the antenatal clinics associated with the study. Subsequent study visits will be conducted at study clinics.

National treatment guidelines stipulate HIV treatment must be initiated promptly in late pregnancy ('same day' in both South Africa and Uganda). Eligibility for DolPHIN-2 will be determined according to patient history, clinical examinations, and laboratory tests for Hb, eGFR and liver function (ALT and bilirubin). Practically, clinical eligibility (history and examination) can be confirmed at screening, but it will not be possible to obtain same day results to confirm laboratory eligibility.

We are therefore proposing to enrol participants based on fulfilling clinical eligibility criteria (history and physical examination), **with a review within 7 days to confirm laboratory eligibility** (eGFR ≥50 mL/min, Hb ≥8g/dL, ALT < 5x ULN, or ALT 3-5 x ULN with bilirubin<2xULN (<35% direct)).

The justification for initial enrolment based on clinical eligibility only is based on the following:

6.4.1 Same-day ART without laboratory results is safe and ethical

- Under the National Treatment Programmes for Uganda and S Africa, same day ART initiation is undertaken based on clinical criteria alone- ie standard of care requires ART initiation ahead of laboratory test results being available

- Same-day initiation of ART (based on clinical eligibility criteria and without available laboratory results) has been shown to be safe and result in faster virological suppression in non-pregnant adults across diverse healthcare settings, such as the RAPID ART Programme in San Francisco (Pilcher, Ospina-Norvell *et al*. 2017), and the RapIT RCT in S Africa randomising 187 newly diagnosed participants to initiate treatment at first visit (Rosen, Maskew *et al*. 2016). Safety and effectiveness has also been retrospectively evaluated in pregnant mothers in the Malawian national treatment programme (N=10,528) (Chan, Kanike *et al*. 2016).

- In the largest head-to-head RCT of DTG versus EFV (SINGLE, N=833), the frequency of laboratory adverse events was not greater with DTG than with EFV. Specifically, with regard to screening investigations, abnormality of ALT, Hb and creatinine were not more frequent, and robust safety data are available.

Table 3 Lab safety events from SINGLE (Walmsley, Antela et al. 2013).

		Grade 1	Grade 2	Grade 3	Grade 4	TOTAL
Any lab event	DTG (414)	10 (2%)	10 (2%)	2 (<1%)	0	22 (5%)
	EFV (419)	8 (2%)	11 (3%)	6 (1%)	2 (<1%)	27 (6%)

Anaemia	DTG (414)	1	0	0	0	1 (<1%)
	EFV (419)	3	0	1	0	4 (<1%)
ALT	DTG (414)	1	1	0	0	2 (<1%)
	EFV (419)	0	3	0	0	3 (<1%)
Creatinine	DTG (414)	0	1	0	0	1 (<1%)
	EFV (419)	0	1	0	0	1 (<1%)

6.4.2 Same-day ART with EFV, followed by a subsequent switch to DTG may contaminate the DTG arm

- Starting mothers on standard of care (EFV-based regimen) carries a risk of a negative drug-drug interaction, as EFV is an enzyme inducer and will lower concentrations of DTG, decreasing overall exposure by 57% [SMPC].

6.4.3 Safeguards have been established to ensure patient safety, safe transfer of participants who subsequently withdraw due to not meeting laboratory inclusion criteria, and statistical handling of withdrawals.

- At the first interim analysis, and on all subsequent analyses, the IDSMB will evaluate any SAEs or untoward events relating to enrolment by clinical criteria only, and advise the TSC and TMG of any recommended amendments to the protocol.
- Participants with abnormal laboratory results will be referred to the national treatment programme (as with participant completing DolPHIN-2, they will be personally escorted to clinics which are adjacent to the research clinics). All laboratory results will accompany the patient, and further investigations will be managed by the clinic.
- We have provided counselling services at each trial site, and these services are also available to people who have been screened for DolPHIN-2, but who are not enrolled, or who do not continue in trial.
- Similar thresholds for laboratory criteria triggering study withdrawal will be applied across both arms, which will ensure that participants who are subsequently withdrawn are approximately balanced across both arms. These individuals will be transferred onto national treatment programme.
- All participants who exit at this stage will be replaced (through blinded randomisation). Participants who are withdrawn for any other reason will not be replaced.

Globally there is a move to implement DTG as a WHO first line regimen (Vitoria, Ford *et al*. 2017)– this is notwithstanding the current lack of data about its use in pregnancy. In 2016, Botswana's 'Treat All' programme implemented universal DTG, replacing EFV. Other 'early implementer' countries are set to follow. Following the potential safety issue affecting women using DTG at the time of conception, WHO advised that pregnant women taking DTG should

not stop; ARV therapy for women of childbearing age should be based on drugs for which adequate efficacy and safety data are available; and if other ARVs cannot be used DTG may be considered in cases where consistent contraception can be assured.

Currently, in Uganda and S Africa there remains equipoise over use of EFV versus DTG in pregnant women. Clearly, this is a scenario which requires regular review – a task which will be undertaken by our TSC and IDSMB. Registry data have also accrued on pregnancy outcomes with DTG, which although lacking the detail, safety assessments and precise characterisation of DolPHIN-2, have also been used to support early implementer policies. Thus, we have considered that it is not unreasonable, and ethically justified to randomise mothers to a DTG-containing arm under a 'same-day' ART initiation programme.

6.5 Withdrawal of participants

A participant is free to withdraw from the study at any time; continued participation in the study is voluntary. In addition, the Investigator may decide, for reasons of medical prudence, to withdraw a participant. Safety bloods and HIV VL results will be communicated to the clinicians responsible for routine care of the participant, and they can choose to withdraw a patient from the study at any time. If a participant discontinues study medication dosing, every attempt should be made to keep the participant in the study and continue to perform the required study-related safety procedures. If this is not possible or acceptable to the participant or Investigator, the participant may be withdrawn from the study.

Study medication may be discontinued in the following instances:

- If the participant withdraws her consent.
- If screening results show that eligibility criteria are not met
- If the Investigator considers in the interest of the participant (i.e. intercurrent illness, unacceptable toxicity) that it is best for her to withdraw her consent.
- The participant's clinician considers it in their best interests
- The participant fails to comply with the protocol requirements or fails to cooperate with the Investigator.
- The participant experiences a liver event, hypersensitivity reaction or severe skin rash
- If the infant develops a toxicity considered to be related to breastmilk exposure to DTG
- Development of active tuberculosis
- New severe, life-threatening opportunistic infection or medical condition
- Severe psychological illness or cognitive deficit such that ongoing consent to participate is in doubt, or difficult to ascertain
- In mothers who have delivered, occurrence of a subsequent pregnancy
- Unwillingness to use effective contraception postpartum
- Intolerance to both EFV and DTG

The date and reasons for the withdrawal will be clearly stated on the participant's CRF. Full detail on exit procedures is in Section 8.14 and in the DolPHIN-2 Treatment Transition and Access Policy.

6.6 Replacements

Participants who discontinue due to ineligibility resulting from laboratory investigation results will be replaced, but participants who discontinue participation for other reasons will not be replaced. For participants found to be ineligible, all stored specimens will be discarded.

6.7 TRIO EXTENSION PARTICIPANT POPULATION

The study population will be recruited from the recruited participants for the DolPHIN 2 study. The total potential population is the number of patients recruited to study (268).

Number of Participants

The number of participants will be determined by which individuals in the DolPHIN 2 study consent to further follow up as part of this extension. Any patient who did not finish the study, or was lost to follow up, will be invited to consent to the TRIO extension. Any patient who withdrew consent from the DolPHIN 2 study will not be invited to participate.

7. STUDY TREATMENTS

7.1 Treatment Plans and Regimens

- Dolutegravir group (DTG+2 NRTIs) – to make best comparison with standard of care, these NRTIs should be those recommended by national policy.
  Participants randomized to the study drug will be commenced on an antiretroviral regimen comprising DTG 50mg once daily in combination with 2 NRTIs
- Standard of care (SoC)/ EFV group
  Participants randomized to receive standard of care will receive the currently used antiretroviral regimens in keeping with national policy (EFV + 2NRTIs at both study sites).
- Backbone 2 NRTIs at treating physician's discretion, according to National guidelines. Those with positive HBsAg will routinely be offered TDF and 3TC (this is in fact the standard of care NRTI backbone in both Uganda and South Africa)

7.2 Dolutegravir

DTG is licensed for HIV-1 treatment in adults and children over 12 years of age and weighing > 40kg. Appendix A contains detailed safety Information data for Tivicay (ViiV 2013). DTG is available as 50mg tablets which do not require any special storage conditions. Peak concentrations of DTG are achieved 2.5 – 4.5 hours post dose, and the terminal half-life of 15 hours supports once daily dosing (Castellino, Moss *et al.* 2013). DTG was licensed in South Africa in 2014 and in Uganda in December 2015.

The safety assessment of DTG in HIV-1 infected treatment naïve participants is based on the analyses of 48 week data from two ongoing, international, multicentre double blind trials,

SPRING-2 (ING113086) (van Lunzen, Maggiolo *et al*. 2012, Raffi, Rachlis *et al*. 2013) and SINGLE (ING114467) (Walmsley, Antela *et al*. 2012) in which a total of 1655 participant received DTG, in combination with 2 NRTIs. The nature and frequency of adverse events are summarized in Table 3.

In SPRING-2, involving 822 participants across 100 sites in North America, Europe and Australia, DTG (co-formulated with TDF/FTC or ABC/3TC) compared favourably with RAL both in terms of virological non-inferiority and with a comparable adverse event profile. The most commonly reported adverse effects were minor gastrointestinal disturbance (nausea or diarrhoea), headache and nasopharyngitis. In both RAL and DTG study arms, 2% of participants suffered adverse events necessitating a change in regimen. Three participant receiving DTG suffered a serious adverse event, namely aphasia, diarrhoea and hepatitis, each occurring in a single participant; no deaths related to the study drug (Raffi, Rachlis *et al.* 2013).Table 2: Treatment-emergent adverse drug reactions of at least moderate intensity (Grades 2 to 4) and ≥2% frequency in treatment-naïve participants in SPRING-2 and SINGLE trials (week 48 analysis).

Table 4 indicates the proportions of adverse events seen in these clinical trials. Recent evidence indicates that in clinical practice DTG discontinuations due to CNS effects including insomnia, sleep disturbance, anxiety and depression affected 5-10% of individuals (de Boer, van den Berk *et al*. 2016). In DolPHIN-2 we will be specifically monitoring participants for CNS toxicities as detailed in Section 4.2.

Table 4 Frequency of adverse events in SPRING-3 and SINGLE studies

	SPRING-2		SINGLE	
System Organ Class/ Preferred Term	TIVICAY 50 mg Once Daily + 2 NRTIs (N = 403)	Raltegravir 400 mg Twice Daily + 2 NRTIs (N = 405)	TIVICAY 50 mg + EPZICOM Once Daily (N = 414)	ATRIPLA Once Daily (N = 419)
Psychiatric				
Insomnia	<1%	<1%	3%	2%
Abnormal dreams	<1%	<1%	<1%	2%
Nervous System				
Dizziness	<1%	<1%	<1%	5%
Headache	<1%	<1%	2%	2%
Gastrointestinal				
Nausea	1%	1%	<1%	3%
Diarrhea	<1%	<1%	<1%	2%
Skin and Subcutaneous Tissue				
Rash[a]	0	<1%	<1%	6%
Ear and Labyrinth				
Vertigo	0	<1%	0	2%

7.3 Prior and Concomitant Medication

Participants should continue to take their prescribed medication, unless it is specifically contraindicated to take this medication with DTG as indicated in the prescribing information for DTG. Both prior and concomitant medication prescribed during the course of the study will

be recorded on the CRF.

- Participants who are concurrently prescribed medication containing divalent cations, such as ferrous sulphate or antacids, will be given specific advice to ingest DTG either two hours before or six hours after these medications.
- Participants receiving DTG should not receive doses of metformin >2g/day, especially if eGFR is < 60 mL/min
- A list of contraindicated medications is provided under 'Exclusion Criteria' above.

7.4 Infant Nevirapine

Under routine PMTCT care, infants will be administered NVP syrup (with or without ZDV/3TC) for the first four to twelve weeks of life (depending on risk stratification and national guidelines). These infants will potentially be exposed to a small dose of DTG via breastfeeding (data regarding the breastmilk elimination of DTG will become available from the DolPHIN-1 study). As DTG is a substrate of UGT1A1 with some contribution from CYP3A, NVP has the potential to decrease the levels of DTG, but this is unlikely to be of clinical significance as the infant DTG is not intended to be of therapeutic benefit.

7.5 TRIO EXTENSION STUDY TREATMENTS

Participants will not be randomised to an additional study treatment as part of the TRIO Extension. South African participants will continue with either DTG or EFV based on their original randomised arm, or if they swapped treatment during or after the study, to their current treatment regime.

Uganda patients will all have their ARVs supplied as part of the national treatment program.

8. STUDY SCHEDULES AND PROCEDURES

The schedule of assessments for each participant enrolled in the study is presented in the study flowchart and detailed in the text below. The shaded section illustrates how proposed study visits relate to the scheduled antenatal and postnatal care of the mother and infant; the study design has taken into consideration the logistical challenges of mothers attending clinic appointments in the early neonatal period and has sought to overlap with the scheduled visits occurring under standard of care.

Study Schedule (main study)

Study Procedure	**Antenatal**				**Delivery**	**Postnatal**					**Exit**
Visit Number	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	
Time	**Screening + Enrolment**	**1w**	**4w**	**36w gest**	**(Estimated) (+14 days)**	**6w**	**12w**	**24w**	**48w**	**72w**	**Any time**
Routine Obstetric Care	ANC V2 (24-28 weeks)	ANC V3 (32-36 weeks)				Postnatal visit ~ 6w					
Informed Consent	X										
Affirmation of consent for continued participation					X						
Screening	X										
Maternal clinical assessment ˣ	X	X	X	X	X	X	X	X	X	X	X
Dispense ART (SoC or DTG)	X	X	X		X	X	X	X	X		
Ultrasound scan (where possible)	X	X	X								
Randomisation 1:1 DTG vs SoC ¥	X										
Allocate Study Treatment	X										
Safety questionnaire - mother	X	X	X	X	X	X	X	X	X	X	X
Maternal safety bloods ~	X	X	X	X		X	X	X	X	X	X
Maternal blood for genomics		X									
Store plasma (EDTA tube, 2 x 1.5 mL aliquots)	X	X	X	X	X	X	X	X	X	X	X
Store serum £	X				X						
Maternal plasma HIV viral load	X	X	X	X	X	X	X	X	X	X	
Paired cord and maternal blood (+store sample)					X						
Collect section of placenta (if possible)					X						
Breast milk HIV viral load						X	X	X	X #		
Store BM						X	X	X	X #		
CD4 count	X							X	X		
Cervico-vaginal secretions sample	X		X								
Confirm contraception					X	X	X	X	X	X	
Preganncy test							X	X	X	X	
Plasma for contraception						X	X	X	X	X	
EPDS and HAD	X	X	X	X		X	X	X	X	X	
Sleep questionnaire		X	X					X			
Social economic questionnaire						X	X +	X +	X +		
Quality of life questionnaires ^		X						X	X		
Hyperglycaemia store samples (serum & plasma)									X	X	
					Infant Procedures						
Safety questionnaire - infant						X	X	X	X	X	X
Child development tool								X	X	X	
Infant clinical assessment					X	X	X	X	X	X	X
Infant blood glucose					X	X					
Infant safety bloods *						X					
Store infant blood (DBS and plasma)					X <	X					
Store infant blood (HIV PCR)							X	X	X		
Infant throat swab & stool swab $						X					
Infant HIV PCR					X	X				X	

ANC =Antenatal Clinic; EPDS = Edinburgh Postnatal Depression Scale; HAD = Hospital Anxiety and Depression Score

ˣ *Clinical assessment – clinical questionnaire, blood pressure, heart rate, urinalysis for protein, blood and glucose*

~ *Maternal safety bloods: FBC, creatinine, ALT, bilirubin, creatine phosphokinase, plasma for storage, fasting glucose, HbA1C, magnesium at Visits 9 and 10 only. If participant has not fasted at Visit 9 or 10 a random glucose sample will be taken*

* *Infant safety bloods: glucose, creatinine, ALT, bilirubin*

\# *To be collected unless weaning has occurred*

^ *Including MOS-HIV, EQ-5D-5L*

$ *Only in infants if mother has given cervico-vaginal secretion sample*

< *Infant plasma sample only at Visit 5*

£ *For folate and magnesium testing if required*

\+ *Only if missed at visit 6 and not done in prior visit*

Screening bloods = FBC, creatinine, ALT, bilirubin, creatine phosphokinase, eGFR, sodium, potassium, urea, HIV 1 & 2 antibodies, CD4 count, HIV viral load, folate levels

Store infant blood sample tests to include HIV PCR, ARV drug levels

8.1 Screening

Screening will take place in the antenatal clinics at the study sites. Any newly diagnosed patient will be counselled regarding their HIV diagnosis and the need to commence anti-retroviral therapy for life, both to reduce the risk of viral transmission to the infant, and for the health of the mother. Counselling will be undertaken by trained nurses and counsellors working in the ART clinic. National policy in both Uganda and South Africa states that women diagnosed with HIV in pregnancy should commence ART on the same day in order to increase the likelihood of an undetectable viral load by the time of delivery. Specialised counselling is necessary since it is also important to ensure the mother is engaged with treatment, understands the diagnosis, the need for treatment, the importance of robust adherence to therapy and the need to attend for follow-up visits at the appointed time. All participants screened, irrespective of whether they are enrolled, will be registered with the National Treatment Programme.

Each potentially eligible participant must sign an Informed Consent Form prior to the conduct of any screening procedures. Participants will be given the opportunity to ask any questions regarding the trial at this stage. Screening evaluations will be used to determine the eligibility of each candidate for study enrolment. Each participant being screened for study enrolment evaluation will be assigned a unique screening participant number. Only one unique identifier will be allocated to a participant.

The screening visit will evaluate:

- Demographic details including age, gender, ethnicity
- History of any previous ART use, including previous PMTCT options or default from first-line therapy. Any previous viral load testing should be recorded.
- Full medical, drug (including contraceptives, herbal/traditional and alcohol/recreational drug use prior to and during this pregnancy) and social history
- Administration of depression scores (Edinburgh Postnatal Depression Scale [EPDS] and Hospital Anxiety and Depression [HAD] Scale)
- Weight, height and vital signs (temperature, blood pressure, heart rate, and respiratory rate)
- Assessment of gestation via LNMP (last normal menstrual period), symphis-fundal height examination and ultrasound scan
- Full blood count with a differential white cell count
- Chemistry panel to include sodium, potassium, urea, creatinine, ALT, bilirubin, creatine phosphokinase and eGFR (calculated from Cockroft-Gault formula)
- HIV-1 and 2 antibodies
- CD4 count
- HIV viral load
- Urinalysis for protein, blood and glucose
- Results of these ‘screening bloods’ will become available within a week from screening, with appropriate clinical care provided, regardless of continued participation.

Where a participant is found to be ineligible due to a laboratory abnormality, she will be referred through best available local practice for clinical input if this is required. Irrespective of whether she requires specialist follow-up, she will offered a follow-up appointment approximately 2 weeks later for clinical review.

Should a participant be ineligible due to evidence of psychiatric morbidity at baseline (including any morbidity detected through HAD and EPDS), she will be referred for formal mental health evaluation and input. At IDI, Kampala, there is a regular mental health clinic within the HIV Prevention, Care and Treatment centre. At Gugulethu the participant would be assessed by public sector social worker and/or community psychiatric nurse and referred to further services as appropriate.

Ultrasound scanning

Where possible, an ultrasound examination will be undertaken at the screening visit to examine for presence of congenital anomalies, and to estimate gestational age. If a medically significant anomaly is detected, consultation will take place between the participant, counsellors, clinicians and the study team regarding subsequent management. The participant may elect to continue in the trial, in which case close communication between clinicians and trial team will continue.

Where an ultrasound examination is not possible at screening, this will be carried out as soon as feasible, and within 2 weeks of enrolment. Further ultrasound scans will be offered at 1 and 4 weeks following ART commencement to assess fetal growth and placental blood flow.

8.2 Visit 1: Enrolment, randomisation and drug dispensing

8.2.1Enrolment and Randomisation

National policy in both Uganda and South Africa states that in pregnant women, ART should commence on the day of HIV diagnosis or first antenatal care visit. Therefore participants who, following screening, meet all eligibility criteria other than laboratory investigations will be enrolled on the same day and randomised to commence either DTG+2NRTIs or EFV+2NRTIs on a 1:1 ratio prior to the availability of blood results (See Section 6.4 for Ethical Justification of this). Participants will be assigned to study treatment in accordance with the computer generated randomization schedule. The randomization schedule will be generated using permuted block randomisation method stratified by country. SAS Proc Plan will be used to generate the randomisation code by the trial statistician at Tropical Clinical Trials Unit (tCTU) at LSTM.

At randomisation, a single communication will be made to both the tCTU and the CRO to notify that a participant has been randomised and drug has been dispensed. This will occur within 24h of drug dispensing.

Following enrolment, subsequent study visits will take place at study clinics and/or antenatal clinics associated with the study, and there will be close liaison with the antenatal teams regarding the clinical care of the participant.

8.2.2 Drug Dispensing

Once the study team is satisfied that the participant understands her diagnosis, the need for treatment, and the importance of adherence, she will be provided with a 1 week supply of DTG+2NRTI or EFV-based SoC and given both verbal and written instructions regarding their administration. Maternal blood will also be taken for genetic testing for polymorphisms associated with drug efficacy or toxicity, and a sample of cervicovaginal fluid will be collected. She will be given a contact number whereby she can raise any questions with a member of the study team at any time. In the event of no further problems, a follow-up appointment shall be made for the following week. Drug dispensing will then take place at each subsequent visit whilst the participant remains in the trial.

8.2.3 Visits 2 and 3

Participants recruited in very late pregnancy may deliver before completing visits 2 and 3. Participants may continue in the study as long as eligibility confirmation has taken place.

8.3 Visit 2, up to Day 7: Eligibility confirmation

Eligibility criteria will be reviewed:

a) Participants who have laboratory results which render them ineligible will be withdrawn from the study, and transferred to the National Programme to receive standard of care ART.
b) Participants who remain eligible will continue their allocated treatment and subsequent study visits as per protocol

For participants who remain in the study, clinical assessment will be undertaken, involving questions regarding the occurrence of any adverse events or difficulties in taking the medication, administration of safety questionnaires, a physical examination, and performance of the panel of 'safety' bloods. A plasma sample will be stored (6mL EDTA blood, allowing storage of 2 x 1.5 mL aliquots). Maternal blood will be collected to measure HIV viral load and for genomics. The EPDS, HAD and quality of life questionnaires (including MOS-HIV) will be administered. The Pittsburgh sleep questionnaire will be administered. A repeat ultrasound will be performed if possible. If uneventful, a follow-up appointment shall be made for three weeks later.

8.4 Visit 3, Day 28 (tolerance ± 14 days)

Clinical assessment will be undertaken, involving questions regarding the occurrence of any adverse events or difficulties in taking the medication, administration of safety questionnaires (Appendix 2) in addition to the EPDS (Appendix 3), the HAD (Appendix 4) (+/- other instruments), a physical examination, and performance of the panel of 'safety' bloods. The Pittsburgh sleep questionnaire will be administered. Maternal HIV viral load will be measured and plasma will be stored. A sample of cervical secretions will be obtained. A repeat ultrasound will be performed if possible.

If there are no complications, the mother shall be reminded of plans for labour and delivery,

and provided with written information to take with her to the labour ward containing details of the study and how to contact the study team, in addition to the information she has already received. If the mother is less than 34 weeks gestation at Visit 3, she will be invited to attend a study visit when she reaches 36 weeks gestation.

8.5 Visit 4 (if required): 36 weeks gestation (tolerance ± 14 days)

For women enrolled at less than 32 weeks gestation, a clinical assessment will be undertaken, involving questions regarding the occurrence of any adverse events or difficulties in taking the medication, administration of safety questionnaires, (Appendix 1) in addition to the EPDS (Appendix 2), the HAD (Appendix 3) (+/- other instruments), a physical examination, and performance of the panel of 'safety' bloods. Maternal HIV viral load will be measured and plasma will be stored. If there are no complications, the mother shall be reminded of plans for labour and delivery, and provided with written information to take with her to the labour ward containing details of the study and how to contact the study team, in addition to the information she has already received.

8.6 Adherence Counselling

All participants will be offered an additional counselling session. This optional visit will be within 1 week of estimated delivery date. Counselling will focus on the importance of adherence to medication post-partum.

8.7 Visit 5, Delivery (tolerance for blood sampling + 14 days)

At delivery, paired cord and maternal blood (plasma) and placenta will be collected, wherever possible, and stored for PK analysis and maternal HIV viral load testing if required. These specimens will be collected by labour ward staff at participating delivery facilities who have been trained in standard operating procedures for the collection of these specimens.

Following delivery (up to within 14 days of delivery), timed to co-incide with their routine-mother infant clinic visit for cord care and neonatal review (standard practice in SA), participants will be asked to attend a study visit. At this visit, the participant will be asked to affirm their consent to the study/ confirm that she gives consent to remain in the study and for infant assessments and sampling to take place as per protocol. If she provides affirmation of informed consent, study procedures will continue, whereas if she does not, she will be withdrawn from the study. If withdrawal occurs the following assessments will not be undertaken instead section 8.14 will be followed.

Maternal clinical assessment and safety questionnaire will be undertaken. Maternal blood will be taken to measure HIV VL. Maternal plasma will be stored.

Infant assessment: The infant will be assessed as follows:

- HIV DNA PCR, if no testing conducted from birth
- Mode of delivery, length of rupture of membranes, and any complications, since can alter transmission risk.

- Gestational age
- Neonatal length and weight
- Neonatal head circumference

Absence or presence of congenital malformations, with description of malformations where present. Surface inspection (with more detailed examination as guided by symptoms or signs) (Mehta, Clerk *et al.* 2012) using the system outlined in the WHO training resources http://www.who.int/tdr/news/2012/pregnancy_registry_protocol/en/ and by Ballard Score (Appendix 6 Liverpool Causality Assessment Tool for Adverse Drug Reactions)

- APGAR scores
- Ballard score
- Presence or absence of intrauterine growth retardation (if data available)
- Plasma glucose for hypoglycaemia
- Plasma stored for pharmacokinetic analysis

As study personnel will not be present at every delivery, the delivery visit may occur up to 14 days after delivery. Data will therefore be collected at both the delivery visit and also from retrospective review of the delivery medical notes. Mode of data collection will be captured on the CRF.

After the delivery study visit, the participant will be given an appointment to attend the research facility in approximately six weeks. In the South African centre, where possible, participation will be documented in the infant 'road to health' booklet. If it is impossible to obtain maternal blood and blood for infant PCR at the time of delivery, these samples will be collected within 14 days. If the infant is HIV positive, they will be referred through local channels for initiation of ART and an infant viral load test will be taken.

If additional bloods are undertaken during the study for clinical reasons, the results of these shall be noted.

All women will be offered contraceptive advice. The potential risks and benefits of different contraceptive methods and of undergoing a subsequent pregnancy will be discussed. For women on DTG, the current evidence regarding DTG exposure in pregnancy will be summarised. Women on DTG who decline effective contraception will be switched from DTG to SoC. They will remain in the study and will be followed up for safety evaluation. All contraceptive use will be recorded on the concomitant medications CRF and contraception CRF.

Effective forms of contraception are:

- Abstinence from penile-vaginal intercourse on a long term and persistent basis
- Contraceptive subdermal implant (DTG only)
- Intrauterine device or intrauterine system

- Combined estrogen and progesterone oral contraceptive (DTG only)
- Injectable progesterone
- Contraceptive vaginal ring
- Percutaneous contraceptive patches (DTG only)
- Female or male partner sterilisation prior to study visit
- Consistent and correct use of barrier methods (male/female condoms) preferably with hormonal implants or IUD, in line with country-specific recommendations

8.8 Visit 6, 6 weeks postpartum (tolerance ± 14 days)

This study visit will coincide with the initial post-delivery assessment which will occur under SoC. This visit will focus on safety endpoints, including maternal clinical assessment, bloods and a neonatal review of the infant by the study doctor, using the WHO recommended surface examination checklist (Mehta, Clerk *et al*. 2012). Both maternal and infant safety questionnaires (Appendix 1) will be administered in addition to the Edinburgh Postnatal Depression Scale, the Hospital Anxiety and Depression Scale (+/- other instruments) and maternal and infant safety bloods will be performed, including infant blood glucose and a dried blood spot and plasma sample from the infant will be stored. Infant blood will be collected for HIV PCR. An optional infant throat swab and stool swab will be obtained if a maternal cervico-vaginal secretions sample was previously collected (Section 12.3). Maternal HIV viral load will be measured, with a sample stored for potential resistance profiling. A 5 mL sample of manually expressed breast milk will be obtained to measure HIV viral load, and the remaining sample will be stored. The time of the last ARV ingestion will be noted. A social economic information questionnaire will be administered (Appendix 11 Social Economic Questionnaire).

If detailed infant examination was not performed by the study team at delivery, it will be performed at this study visit. In the South African centre, if not already done postpartum, participation in the study will be documented in the infant 'Road to Health' booklet.

Women will also be provided with routine postpartum care including contraceptive advice. The potential risks and benefits of different contraceptive methods and of undergoing a subsequent pregnancy will be discussed. For women on DTG, the current evidence regarding DTG exposure in pregnancy will be summarised. Women on DTG who decline effective contraception or who are pregnant will be switched from DTG to SoC. They will remain in the study and will be followed up for safety evaluation. All contraceptive use will be recorded on the concomitant medications CRF and contraception CRF.

8.9 Visit 7, 12 weeks postpartum (tolerance ± 14 days)

A full clinical assessment of mother and infant, including maternal and infant safety questionnaires, EPDS and HAD and sampling of maternal safety bloods, maternal HIV viral load (in blood and breast milk) will be performed. Maternal blood and breast milk and infant plasma will be stored. A social economic information questionnaire should be administered (Appendix 11 Social Economic Questionnaire) if previously missed on visit 6.

Women will be provided with contraceptive advice. The potential risks and benefits of different contraceptive methods and of undergoing a subsequent pregnancy will be discussed. For women on DTG, the current evidence regarding DTG exposure in pregnancy will be summarised. A pregnancy test will be performed. Participants who decline a pregnancy test will continue in the trial. Women on DTG who decline effective contraception or who are pregnant will be switched from DTG to SoC. They will remain in the study and will be followed up for safety evaluation. All contraceptive use will be recorded on the concomitant medications CRF and contraception CRF.

8.10 Visit 8, 24 weeks postpartum (tolerance ± 28 days)

A full clinical assessment of mother and infant, including maternal and infant safety questionnaires, EPDS and HAD, and maternal sampling of safety bloods, maternal HIV viral load (in blood and breast milk) and CD4 count will be performed. The Pittsburgh sleep questionnaire will be administered. Maternal plasma and breast milk and infant plasma will be stored. Infant development will be assessed using the infant gross motor screening test (IGMST) (Appendix 4). Quality of life questionnaires including MOS-HIV will be administered. A social economic information questionnaire should be administered (Appendix 11 Social Economic Questionnaire) if previously missed on visit 6.

Women will be provided with contraceptive advice. The potential risks and benefits of different contraceptive methods and of undergoing a subsequent pregnancy will be discussed. For women on DTG, the current evidence regarding DTG exposure in pregnancy will be summarised. A pregnancy test will be performed. Participants who decline a pregnancy test will continue in the trial. Women on DTG who decline effective contraception or who are pregnant will be switched from DTG to SoC. They will remain in the study and will be followed up for safety evaluation. All contraceptive use will be recorded on the concomitant medications CRF and contraception CRF.

8.11 Visit 9, 48 weeks postpartum (tolerance ± 28 days):

A full clinical assessment of mother and infant, including maternal and infant safety questionnaires, EPDS and HAD, and maternal sampling of safety bloods, maternal HIV viral load and CD4 count will be performed. Maternal plasma and infant plasma will be stored. Maternal breast milk for HIV viral load and storage will be collected unless the weaning has occurred. Infant development will be assessed using the infant gross motor screening test (IGMST). Quality of life questionnaires including MOS-HIV will be administered. A social economic information questionnaire should be administered (Appendix 11 Social Economic Questionnaire) if previously missed on visit 6.

Women will be provided with contraceptive advice. The potential risks and benefits of different contraceptive methods and of undergoing a subsequent pregnancy will be discussed. For women on DTG, the current evidence regarding DTG exposure in pregnancy will be

summarised. A pregnancy test will be performed. Participants who decline a pregnancy test will continue in the trial. Women on DTG who decline effective contraception or who are pregnant will be switched from DTG to SoC. They will remain in the study and will be followed up for safety evaluation. All contraceptive use will be recorded on the concomitant medications CRF and contraception CRF.

8.12 Visit 10, 72 weeks postpartum (tolerance +/- 28 days) End of Study

A final follow-up appointment will take place at 72 weeks for mother and infant clinical assessments, including maternal and infant safety questionnaires, EPDS and HAD, maternal safety bloods, maternal HIV viral load and infant HIV PCR. Maternal plasma will be stored. Infant development will be assessed using the infant gross motor screening test (IGMST). Participants will be exited from the study at 72 weeks post-partum. See Section 8.18 for procedures required for study exit.

Women will be provided with contraceptive advice. The potential risks and benefits of different contraceptive methods and of undergoing a subsequent pregnancy will be discussed. For women on DTG, the current evidence regarding DTG exposure in pregnancy will be summarised. A pregnancy test will be performed. Participants who decline a pregnancy test will continue in the trial. All contraceptive use will be recorded on the concomitant medications CRF and contraception CRF.

If no complications are identified, the End of Study case record forms will be completed. Transitional access will be discussed and the mother will be transitioned to the National Treatment Programme (NTP) for her HIV management – these processes are detailed in Section 8.18 and Section 9, respectively.
Propose to attend the TRIO visits below

8.12.1 Drug dispensing visits

Clinical visits for drug dispensing and clinical follow-up in accordance with National Policy will take place between the specified Study Visits and will be led by members of the study team.

8.13 Clinical Assessment of Mothers and Infants

See sections 10.1 and 10.2.

8.14 Withdrawal from the study

Participants who are withdrawn from the study due to failure to meet eligibility criteria on the basis of laboratory results will be transitioned to the NTP to receive standard of care antiretroviral therapy. These participants will be replaced and will not be eligible for transitional access to study drug (Section 9).

Participants withdrawing for any other reason will not be replaced.

For early study termination, the mother (and infant), the following evaluations will be performed:

- Vital signs (temperature, blood pressure, and heart rate)
- Haematology with a differential white blood cell count
- ALT, bilirubin, creatinine, CK
- Review of Adverse Events
- Concomitant medications review

The reason for the early termination of the participant should be clearly documented on the participant's CRF. The participant will not then attend further follow up visits unless deemed necessary in the opinion of the investigator (e.g. due to adverse event or to determine pregnancy outcome status if still pregnant at the time of early termination).

8.15 Treatment 'failure', and switching therapy

Switching NRTI backbone for intolerance or toxicity will be recorded, but not constitute a study endpoint. Participants may switch study drug (DTG or EFV) for a number of reasons:

8.15.1 Development of severe adverse events,

Either in the form of meeting liver stopping criteria (see 11.3.1) or other severe adverse reaction

8.15.2 Development of any adverse effects or intolerance

i. Any participant who is intolerant of either EFV or DTG will exit DolPHIN2 and transition to the NTP where they will continue to be followed up for safety endpoints and access second-line agents in line with national treatment guidelines.
ii. Participants who develop toxicity/intolerance to other agents within the regimen will be offered alternative nucleoside drugs. Participants will remain under follow-up in DolPHIN-2 and included in both 'intention-to-treat' and 'as-treated' analyses.

8.15.3 Development of virological failure

Antepartum: it is unlikely that participants initiating ART within the eligible period (28w-term) will all have undetectable viral loads at delivery regardless of regimen, and it is therefore difficult to define a 'failing' regimen at this point. Frequent viral load tracking is routinely undertaken. Therefore we do not envisage treatment switches for 'virological failure' prior to delivery.

Post-partum: Failure to achieve a viral load <50 copies by and including 24w post-partum ('virological non-responder'), or virological responders who subsequently rebound with two consecutive viral loads >1000 copies/mL ('virological rebound') will be classified as 'virological failure'.

i. Any participant who fails either EFV or DTG, they and their infants will remain in the study as follows:
   Mother: Continue trial follow-up and safety reporting. Referred for urgent counselling and continue dispensation of the allocated treatment with delayed viral load assessment

(approximately 1 month afterwards). The participant should have a local HIV resistance test (new clinical sample) and fresh breast milk collected for central laboratory resistance testing (existing sample collected at each visit). EFV dispensation and DTG dispensation shall continue including 6-month post-trial access period according to study arm unless resistance test results suggest otherwise.

Infant(s): Continue follow-up and safety reporting as normal with infant HIV PCR at every visit as clinically indicated irrespective of protocol specification.

The study will obtain samples for HIV resistance testing (genotyping) through nationally accredited laboratories in each country, which detect common mutations to NNRTIs, NRTIs and protease inhibitors) at the time of virological failure. For women with virologic failure in either arm, the subsequent regimen will be informed by the results of resistance testing.

Study participants are deemed to have exited the study when i) at 196 weeks post-partum or ii) development of severe adverse events, either in the form of meeting liver stopping criteria (see 11.3.1) or other severe adverse reaction, or iii) clinician's discretion, or iv) participant wish to exit study. End of Study case record forms should be completed and the clinical trials unit notified.

8.16 Development of mastitis.

Mastitis has been implicated as a risk factor for HIV transmission through breastfeeding in the pre-Option B+ era. All participants will be asked to prospectively record episodes of mastitis during breastfeeding, and to report this at the next scheduled study visit. Feeding should continue on the unaffected breast. Local breastfeeding guidelines should be followed - in general these recommend frequent expression of milk from the affected breast (which is discarded) and treatment of mastitis according to local guidelines which may involve use of cold compresses, antibiotics in the presence of cellulitis, and pain relief.
If mastitis is present during a scheduled study visit, paired samples from plasma and milk from the affected breast will be collected for viral load measurement.

8.17 Adherence support and Counselling

All participants will be offered counselling at the time of receiving an HIV diagnosis, and as required thereafter, and following ultrasound. Study personnel will be trained to support their clients in HIV as well as obstetric-related issues. Counsellors will be present at study clinics.
Participants who meet the threshold for risk of anxiety or depression will be automatically referred for further assessment and counselling, through the structures indicated in Section 10.1.

8.18 Transition to the National Treatment Programme

To prevent gaps in treatment, all participants referred for screening should be registered (at the time of screening, or shortly thereafter) with the NTP as described in section 8.2. For DolPHIN-2 participants who exit prematurely or who attend at 72w post-partum, arrangements

will be made to transition back into care under the NTP clinic offering antiretroviral therapy. The DolPHIN-2 study sites are in Kampala and Cape Town- in both sites, DolPHIN-2 trial clinics are embedded adjacent to, or within general ART clinics:

i) All participants will be personally escorted (if possible) to the ARV clinic, and re-enrolled onto the NTP register.

ii) Handover documentation based on local templates will include the following information if relevant:

- Relevant medical history. Dates of diagnosis and treatment initiation
- All current and previous ART regimens, and all relevant co-medications
- All relevant clinical events, co-infections and potential drug toxicities
- Any abnormalities in safety or baseline bloods
- Assessment of adherence
- Weaning status of infant
- Partner testing, and notification
- Any confidentiality or disclosure issues

iii) The DolPHIN-2 team will obtain written confirmation of enrolment of the participant into the ARV clinic system wherever possible.

iv) The participant will retain contact details of the DolPHIN-2 trial team, in case of problems encountered in the transition process

8.19 TRIO EXTENSION STUDY SCHEDULES AND PROCEDURES

Patients will re-enter the study at the most appropriate visit, but can be reconsented prior to this visit.

Table 5:

Procedure	Visit 11 96 w pp	Visit 12 120 w pp	Visit 13 144 w pp	Visit 14 168 w pp	Visit 15 192 w pp
MOTHER					
Re-Consent	X*				
Weight	X	X	X	X	X
Vital signs[1]	X	X	X	X	X
Maternal clinical assessments	X	X	X	X	X
Maternal Safety questionnaire	X	X	X	X	X
AE/SAE review[2]	X	X	X	X	X
Concomitant medication[2]	X	X	X	X	X
Drug adherence monitoring	X	X	X	X	X
Maternal HIV viral load	X	X	X	X	X
Maternal CD4 count	X	X	X	X	X

FBC inc differential[3]	X	X	X	X	X
Chemistry bloods[4]	X	X	X	X	X
Plasma sample (PK/resistance testing, contraception & complex biomarkers)	X	X	X	X	X
Serum gel sample (insulin/C-peptide/leptin)[5]	X	X	X	X	X
Fluoride plasma sample (glucose)[5]	X	X	X	X	X
Urine pregnancy test	X	X	X	X	X
HbA1c[6]	X	X	X	X	X
Urinalysis[7]	X	X	X	X	X
Subsequent pregnancy follow up					
Serum HCG	X	X	X	X	X
INFANT					
Vital signs[1]	X	X	X	X	X
Infant safety questionnaire	X	X	X	X	X
Child development tool Bayleys III	-	-	-	X	-
Child development tool CBCL	X	X	X	X	X
HIV -ve infants only					
Infant HIV antibody test	X*				
HIV +ve infants only					
Infant HIV viral load	X	X	X	X	X
Infant CD4 count	X	X	X	X	X
Infant resistance test[8]	X	X	X	X	X

[1]	Blood Pressure, Heart Rate, Respiratory Rate, Temperature
[2]	From date of last study visit
[3]	WBC, Haemoglobin, Platelets, Haematocrit, Neutrophils, Monocytes, Lymphocytes
[4]	Creatinine, Bilirubin, ALT, CK, Urea, Potassium, Sodium, eGFR, hsCRP, LDH, GGT, ALP, Triglycerides, Total Cholesterol, LDL, Fasting plasma glucose, Magnesium, HDL, Total protein, Phosphate (serum), HbA1c
[5]	Hyperglycaemia assessment
[6]	Glycated haemoglobin (IFCC), Glycated haemoglobin (NGSP), estimated average glucose (eAG)
[7]	Urinalysis for protein, blood & glucose
[8]	Resistance test only to be taken if suspected virological failure or at time of suspected MTCT
*	Required for 1st follow up visit only

9. TRANSITIONAL ACCESS TO DTG

The DolPHIN-2 team are committed to good practice in post-trial care, and have negotiated with ViiV to provide access to DTG post-trial for a period of approximately 6 months after

transition to the NTP (see current version of the transitional access plan for full detail). This access to DTG will be offered to mothers in the DTG-containing arm, unless there is previous history of drug intolerance or severe adverse reactions, or in mothers who are pregnant again, or who are receiving TB therapy, or decline effective contraception. All mothers who become pregnant or require TB therapy whilst receiving DTG will discontinue DTG therapy, and switch to an alternative agent in accordance with national guidelines.
Post-trial access to DTG will be made available only to participants recruited into DolPHIN-2 (ie not the clinic population as a whole, or to family members). DTG will be supplied as labelled drug through the usual pharmacy dispensing ARVs to clinic participants. It is anticipated that DTG will become available in the public sector in South Africa and Uganda within the next two to four years, depending in part on DolPHIN-2 findings.

9.1.1 Gugulethu CHC Arrangements

While it is possible that DTG will be available in the public sector before the end of the study, we have consulted with the Gugulethu CHC and provincial health services to confirm an approach to transitioning that is safe in terms of patient care and acceptable to the local health services. Briefly, individual care plans will be made for each participant in conjunction with the Gugulethu ARV service, with whom we maintain excellent working relations. While care plansy will vary by women's circumstances, all participants will be referred back to the NTP as per local guidelines.

9.1.2 IDI Arrangements with National ARV Program

The IDI is accredited by the Uganda Ministry of Health (MoH) as a provider of specialized HIV services. In addition, IDI serves as implementing partner of the MoH for HIV prevention care and treatment in the Kampala and Wakiso regions. The national HIV/STI program has agreed that DolPHIN-2 participant will be transferred back to the national program at the end of the study. Prior to transition, women experiencing adverse events will be managed in the study as indicated in section 10 of the protocol. Unless requested otherwise by the participant, women will continue their care at the IDI. Participants with detectable viral loads will be managed in accordance with Uganda national treatment guidelines (Uganda 2016).

9.2 TRIO EXTENSION ACCESS TO DGT

Dolutegravir will be sourced locally for the study extension. Participants in Uganda and South Africa should be able to access Dolutegravir as part of their countries respective National Treatment Guidelines. Dolutegravir can be sourced for patients in country as a fixed dose combination or as a single tablet with a separate backbone.

If patients have changed their ARV treatment since randomisation it will not be expected that they return to the randomised arm from the original the DolPHIN 2 study.

10.SAFETY MONITORING

Safety assessments will be conducted to characterize the safety and tolerability of DTG administered during pregnancy, including monitoring and recording all AEs and SAEs, regular monitoring of haematology and blood chemistry, physical exams and monitoring and recording birth and maternal outcomes.

10.1 Maternal Assessments

Laboratory baseline screening is outlined in Section 8.2 and includes:

Full blood count (including automated WBC differential, platelets, haemoglobin, haematocrit), creatinine, creatinine phosphokinase, bilirubin, glucose. Glomerular Filtration Rate (GFR) will be estimated. Blood will be stored for maternal plasma HIV viral load and resistance assays. Urinalysis (dipstick) will test for the presence of protein, blood or glucose.

A safety questionnaire will be administered at every study visit. Women will also be asked to report any prescribed, herbal, traditional or over the counter medications which they have taken since the last study visit. Qualitative evidence drawn from similar populations indicates that direct questions about this are valued and will yield more accurate data than less specific, general questions (Allen, Chandler *et al*. 2013, Allen, Gomes *et al*. 2014)

In samples where HIV viral load is measurable due to presumed loss of virological control, resistance profiling will be done.

The EPDS (Appendix 2) and the HAD (Appendix 3) will be administered at the early postnatal visit, with prompt liaison with psychiatric services should a risk of suicidality be identified. At IDI, Kampala, there is a regular mental health clinic within the HIV Prevention, Care and Treatment centre. At Gugulethu the participant would be assessed by public sector social worker and/or community psychiatric nurse and referred to further services as appropriate.

10.2 Infant Assessment

At delivery infants will be evaluated (see section 8.1 and 8.7) to assess the following:

- Mode of delivery, length of rupture of membranes, and any complications, since can alter transmission risk.
- Gestational age
- Neonatal length and weight
- Neonatal head circumference
- Absence or presence of congenital malformations, with description of malformations where present
- APGAR scores
- Ballard score
- Evidence of intrauterine growth retardation (if data available)
- Glucose
- ART drug levels

Follow-up of infants. A formal paediatric assessment will be undertaken at 6 weeks postpartum and at weeks 24 and 48 following delivery, and at any other time point if concerns are noted. Assessments will be performed as per local guidelines including a history and physical examination. Furthermore, we will report on child development using the Infant Gross Motor Screening Tool (IGMST) This tool has been validated in HIV-infected infants and assesses gross motor development at each age (see Appendix 4).

- Anthropometric measures (weight, length, head circumference)
- Feeding
- Safety bloods (see section 8.1) at 6 weeks to measure bilirubin, ALT, creatinine and glucose. An extra sample of blood (0.5mL) will be collected and stored for PK assessment (time of last maternal drug intake, and most recent breast feed, to be recorded)
- If additional bloods are undertaken during the study for clinical reasons, the results of these shall be noted, and an additional 1mL of blood will be collected for PK assessment (time of last maternal drug intake to be recorded) with a maximum of 3 samples over a one year period.
- Developmental screen: Infants are classified according to an age-related score and any "at risk" infant will automatically be referred to a paediatrician.
- Resistance profiling will be performed on any infant who acquires HIV during the study.

Any infant born to mothers in the study who require paediatric care will be given facilitated access to the local standard of care

10.3 Hyperglycaemia assessment

This will be carried out at Visits 9 and 10. Fasted blood samples will be taken to measure glucose, magnesium and HbA1C in real time via local laboratories. Further blood samples will be taken and stored for analysis of c-peptide, insulin and leptin at a central laboratory. Samples will be stored at site and shipped to a central laboratory for analysis of biomarkers to elucidate the mechanism of action. These samples will still be obtained and analysed if the participant is not fasted on the day of collection.

10.4 TRIO EXTENSION SAFETY MONITORING

Safety assessments will be conducted to monitor the safety of patients and their infant at each visit.

Mothers will have the following at all visits
Vital Signs
Maternal clinical assessments
Maternal safety questionnaire

AE/SAE review
Concomitant medication
Maternal HIV viral load & CD4 count
FBC (inc differential)
Serum gel sample (insulin/C-peptide/leptin) – Hyperglycaemia assessment
Fluoride plasma sample (glucose) – Hyperglycaemia assessment
Chemistry bloods
Urinalysis

Infants will have the following at all visits:
Vital Signs
Infant Safety Questionnaire
Child development tools

HIV -ve infants will have a HIV antibody test at re-entry visit/week 96 (visit 11).
HIV +ve infants will have HIV viral load and CD4 count at all visits and a resistance test if virological failure is suspected or MTCT.

For full details of the assessments see table 5 in section 8.19

11.MANAGEMENT OF ADVERSE EVENTS

11.1 Adverse Events

An adverse event (AE) is any untoward medical occurrence in a participant administered a pharmaceutical product and which does not necessarily have a causal relationship with the study treatments. An AE can therefore be any unfavourable and unintended sign (including an abnormal laboratory finding), symptom, or disease temporally associated with the use of a medicinal (investigational) product, whether or not considered related to the medicinal (investigational) product.

AEs observed by the Investigator, or reported by the participant, and any remedial action taken, will be recorded in the participant's CRF and should be verifiable in the participant's notes throughout the study. The nature of each event, time of onset after drug administration, duration and severity will be documented together with the Investigator's opinion of the causal relationship to the treatment.

Procedures such as surgery should not be reported as AEs. However, the medical condition for which the procedure was performed should be reported if it meets the definition of an AE. For example, an acute appendicitis that begins during the AE reporting period should be reported as the adverse event and the resulting appendectomy noted on the CRF. Planned procedures such as surgery planned prior to the participant's enrolment into the study need not be reported as AEs if these are documented as planned at the screening visit.

Clinically significant changes in physical examination and blood safety profiles should also be recorded as AEs. The specific actions for women who are unable to tolerate their prescribed regimen due to AEs are detailed above in Section 8.14.

Severity should be recorded and graded according to the **Division of Aids Table for Grading the Severity of Adult and Paediatric Adverse Events** (Appendix 5).

Note: There is a distinction between the gravity and the intensity of an AE. Severe is a measure of intensity; thus, a severe reaction is not necessarily a serious AE. For example, a headache may be severe in intensity but would not be classified as serious unless it met one of the criteria for serious events.

The relationship to the study drugs of each AE will be assessed using the Liverpool Causality Tool (Appendix 6) with the following definitions:

DEFINITE: distinct temporal relationship with drug treatment. Known reaction to agent or chemical group, or predicted by known pharmacology. Event cannot be explained by participant's clinical state or other factors.
PROBABLE: reasonable temporal relationship with drug treatment. Likely to be known reaction to agent or chemical group, or predicted by known pharmacology. Event cannot easily be explained by participant's clinical state or other factors.
POSSIBLE: reasonable temporal relationship with drug treatment. Event could be explained by participant's clinical state or other factors.
UNLIKELY: poor temporal relationship with drug treatment. Event easily explained by participant's clinical state or other factors.
The definition for unrelated is as below:
UNRELATED: the event occurs prior to dosing. Event or intercurrent illness is due wholly to factors other than drug treatment.

All AEs, however minor, will be documented in the CRF whether or not the Investigator concludes the event to be related to drug treatment.

The AE reporting period will be from the screening visit until the participant's final study visit. In addition, any untoward event that may occur within 30 days of the last prescribed dose of investigational product that the Investigator assesses as possibly, probably or definitely related to the study drug medication should also be reported as an AE.

AEs may be directly observed, reported spontaneously by the participant or by questioning the participant at each study visit.

All AEs should be followed up until they are resolved or the participant's participation in the study ends (i.e. until the final CRF is completed for that participant). In addition, all serious and non-serious AEs assessed by the Investigator as possibly related to the investigational

medication should continue to be followed even after the participant's participation in the study is over. Such events should be followed until resolution, or until no further change can reasonably be expected.

11.2 Serious Adverse Events

A serious adverse event (SAE) is any untoward medical occurrence that at any dose:

i) Results in death
ii) Is life threatening
iii) Requires in patient hospitalisation* or prolongation of existing hospitalisation
iv) Results in persistent or significant disability/ incapacity, or
v) Is a congenital anomaly/ birth defect.
vi) All events of possible drug-induced liver injury with hyperbilirubinemia defined as ALT ≥3xULN and bilirubin ≥2xULN (>35% direct)

*Hospitalisation for purely obstetric indications will not be considered an SAE unless clinical information suggests otherwise. This data will be captured by the study but not reported as an SAE.

NOTE: bilirubin fractionation is performed if testing is available. If testing is unavailable, record presence of detectable urinary bilirubin on dipstick indicating direct bilirubin elevations and suggesting liver injury. If testing is unavailable and a participant meets the criterion of total bilirubin ≥2xULN, then the event is still reported as an SAE. If PT is obtained, include values on the SAE form. PT elevations >1.5 x ULN suggest severe liver injury.

Liver events: Liver event CRFs and liver imaging and/or liver biopsy CRFs (see Section 11.3.1) should be completed and reported to the Principal Investigator within one week of event onset.

Suicidal Ideation or Behaviours: If any participant experiences a possible suicidality-related adverse event (PSRAE) while participating in this study that is considered by the Investigator to meet International Conference on Harmonization (ICH)-E2A definitions for seriousness, the Investigator will collect information using a PSRAE CRF form in addition to reporting the event on a SAE CRF form. A PSRAE may include, but is not limited to, an event that involves suicidal ideation, a preparatory act toward imminent suicidal behaviour, a suicide attempt, or a completed suicide. The investigator will exercise his or her medical and scientific judgment in deciding whether an event is possibly suicide-related. PSRAE forms should be completed and reported to the Principal Investigator within one week of the investigator diagnosing a possible suicidality-related serious adverse event. Study DTG should not be continued after a suicide attempt.

Deaths occurring more than 30 days after the final dose, which are considered to be unrelated to the study medication, should not be reported as a Serious Adverse Event.

11.3 Specific Toxicities/Adverse Event Management

General guidelines for the management of specific toxicities that are considered to be related or possibly related to EFV or DTG are provided below.

Participants who permanently discontinue either drug for reasons of toxicity should be followed weekly until resolution of the AE and encouraged to complete the withdrawal and FU study evaluations as noted above.

11.3.1 Liver chemistry stopping and follow up criteria

Liver chemistry threshold stopping criteria have been designed to assure participant safety and to evaluate liver event aetiology during administration of IP and the follow-up period. IP will be stopped if any of the following liver chemistry criteria are met:

- ALT ≥3xULN **and** bilirubin ≥2xULN (>35% direct bilirubin; bilirubin fractionation required)
    - NOTE: serum bilirubin fractionation should be performed if testing is available. If testing is unavailable, record presence of detectable urinary bilirubin on dipstick, indicating direct bilirubin elevations and suggesting liver injury. If testing is unavailable and a participant meets the criterion of total bilirubin ≥ 2xULN, then the event meets liver stopping criteria;
- ALT ≥8xULN;
- ALT ≥3xULN (if baseline ALT is < ULN) with symptoms or worsening of acute hepatitis or hypersensitivity such as fatigue, nausea, vomiting, right upper quadrant pain or tenderness, fever, rash, or eosinophilia, OR;
- ALT ≥3x baseline ALT with symptoms or worsening of acute hepatitis or hypersensitivity such as fatigue, nausea, vomiting, right upper quadrant pain or tenderness, fever, rash, or eosinophilia;
- ALT ≥5xULN and <8xULN that persists > 2 weeks (with bilirubin <2xULN and no signs or symptoms of acute hepatitis or hypersensitivity);
- ALT ≥5xULN but <8xULN and cannot be monitored weekly for >2 weeks;

Participants who develop ALT ≥5xULN should be followed weekly until resolution or stabilization (ALT <5xULN on 2 consecutive evaluations).

When liver chemistry stopping criterion is met, do the following:

- **Immediately discontinue DTG. Participants should not restart DTG due to the risk of a recurrent reaction. When clinically appropriate, EFV-based ART will be prescribed and the participant will remain in the study for safety follow-up.**
- Evaluate for HELLP Syndrome and pregnancy-related steatosis in participants with ongoing pregnancy.
- Notify the Principal Investigator of the event by telephone within 24 hours of learning its occurrence;
- Events of possible drug-induced liver injury with hyperbilirubinemia defined as ALT ≥3xULN and bilirubin ≥2xULN (>35% direct) will be reported to the Principal Investigator as SAEs using the SAE CRF (see Section 11.3).

- Complete the liver event CRF for all events meeting liver stopping criteria, and report to the Principal Investigator within one week of first becoming aware of the event (see Section 11.3);
- Complete the liver imaging and/or liver biopsy CRFs if these tests are performed, and report to the Principal Investigator (see Section 11.3);
- Perform liver event follow up assessments (described below), and monitor the participant until liver chemistries resolve, stabilize, or return to baseline values as described below;
- Make every reasonable attempt to have participant return to clinic within 24 hours for repeat liver chemistries, liver event follow up assessments (see below), and close monitoring;
- A specialist or hepatology consultation is recommended;
- Monitor participant at least weekly until liver chemistries (ALT, AST, alkaline phosphatase, bilirubin) resolve, stabilize or return to within baseline values.

Consider the following additional tests to further evaluate the liver event:

- Viral hepatitis serology including:
  - Hepatitis A IgM antibody;
  - HBsAg and Hepatitis B Core Antibody (IgM);
  - Hepatitis C RNA;
  - Hepatitis E IgM antibody;
- Cytomegalovirus IgM antibody;
- Epstein-Barr viral capsid antigen IgM antibody (or if unavailable, obtain heterophile antibody or monospot testing);
- Syphilis screening
- Drugs of abuse screen including alcohol
- Serum acetaminophen test (APAP adduct test)
- Serum creatine phosphokinase (CPK) and lactate dehydrogenase (LDH);
- Fractionate bilirubin, if total bilirubin is greater than 1.5xULN;
- Obtain complete blood count with differential to assess eosinophilia;
- Anti-nuclear antibody, anti-smooth muscle antibody, and Type 1 anti-liver kidney microsomal antibodies;
- Liver imaging (ultrasound, magnetic resonance, or computerized tomography) to evaluate liver disease;
- Record the appearance or worsening of clinical symptoms of hepatitis, or hypersensitivity, fatigue, decreased appetite, nausea, vomiting, abdominal pain, jaundice, fever, or rash as relevant on the AE report form;
- Record use of concomitant medications, acetaminophen, herbal remedies, other over the counter medications, or putative hepatotoxins, on the concomitant medications report form. Record alcohol use on the liver event alcohol intake case report form.

11.3.2 Allergic Reaction

Participants may continue DTG for Grade 1 or 2 allergic reactions at the discretion of the

Investigator. The participant should be advised to contact the Investigator immediately if there is any worsening of symptoms or if further systemic signs or symptoms develop. Antihistamines, topical corticosteroids, or antipruritic agents may be prescribed.

Participants with Grade ≥3 allergic reactions that are considered to be possibly or probably related to DTG should permanently discontinue the investigational product regimen and the participant should switch to EFV. Participants should be treated as clinically appropriate and followed until resolution of the AE.

11.3.3 Rash

Mild to moderate rash is an expected adverse reaction for DTG- containing ART. Episodes generally occur within the first ten weeks of treatment, rarely require interruptions or discontinuations of therapy and tend to resolve within two to three weeks. No instances of serious skin reaction, including Stevens-Johnson Syndrome (SJS), Toxic Epidermal Necrolysis (TEN) and erythema multiforme, have been reported for DTG in clinical trials.

Participants with an isolated Grade 1 rash may continue DTG at the Investigator's discretion. The participant should be advised to contact the Investigator immediately if there is any worsening of the rash, if any systemic signs or symptoms worsen, or if mucosal involvement develops. Any participant who presents with a rash should have their liver function tests checked.

Participants may continue DTG for an isolated Grade 2 rash. However, DTG (and all other concurrent medication(s) suspected in the Investigators causality assessment) should be permanently discontinued for any Grade ≥2 rash that is associated with an increase in ALT. The participant should be advised to contact the physician immediately if rash fails to resolve (after more than two weeks), if there is any worsening of the rash, if any systemic signs or allergic symptoms develop, or if mucosal involvement develops.

Participants should permanently discontinue DTG (and all other concurrent medication(s) suspected in the Investigators causality assessment) for an isolated Grade 3 or 4 rash, and the participant should be withdrawn from the study. Participants should be treated as clinically appropriate and followed until resolution of the AE.

The rash and any associated symptoms should be reported as adverse events (see Section 11.3) and appropriate toxicity ratings should be used to grade the events (based on DAIDS toxicity gradings).

If the aetiology of the rash can be definitely diagnosed as being unrelated to DTG and due to a specific medical event or a concomitant non-study medication, routine management should be performed and documentation of the diagnosis provided.

11.3.4 Suicidal Risk Monitoring

Participants with HIV infection may occasionally present with symptoms of depression and/or suicidality (suicidal ideation or behaviour). In addition, there have been some reports of depression, suicidal ideation and behaviour (particularly in participant with a pre-existing history of depression or psychiatric illness) in some participant being treated with integrase inhibitors, including DTG. Therefore, it is appropriate to monitor participants for suicidality before and during treatment.

Participants should be monitored appropriately and observed closely for suicidal ideation and behaviour or any other unusual changes in behaviour at all study visits. It is recommended that the investigator consider mental health consultation or referral for participants who experience signs of suicidal ideation or behaviour. The EPDS (Appendix 2) and the HAD (Appendix 3) will also be administered at baseline and postnatally (see Section 10.1).

If any participant experiences a possible suicidality-related adverse event (PSRAE) while participating in this study that is considered by the Investigator to meet International Conference on Harmonization (ICH)-E2A definitions for seriousness, the Investigator will collect information using a PSRAE CRF form (see Section 11.3).

11.3.5 Decline in Renal Function

Treatment with any fixed dose combination NRTI products containing 3TC, FTC or TDF must be discontinued in any participant developing moderate to severe renal impairment during the study, as indicated by creatinine clearance measuring <50 mL/min via Cockroft-Gault.

11.3.6 Stillbirth

This study will involve women of ≥28 weeks gestation, and all the internationally used definitions of stillbirth have a cut-off below this threshold (Tavares Da Silva, Gonik *et al*. 2016). Where a woman has delivered a stillbirth, if the mother provides consent we will seek to perform a verbal autopsy in attempt to elucidate the cause of fetal demise. Participants will be given the choice whether to remain in the study or not. Close liaison will take place with the NTP clinic to ensure she has access to available sexual and reproductive health and psychological care in addition to receiving ongoing ART.

11.3.7 Pre-term Delivery

Case definitions for pre-term birth will follow those proposed by the Brighton collaboration (Quinn, Munoz *et al*. 2016). Gestational maturity will be assessed at screening using the best available estimates. This will include LNMP, ultrasound scanning and symphysis fundal height at antenatal visits. In late pregnancy, none of these are ideal. However, pre-term delivery has been associated with both HIV and antiretroviral regimens and is therefore an important outcome to capture. At delivery visit, the Ballard score will be performed to assess maturity. Where there is uncertainty or a major discrepancy (defined as a difference in gestational age that would result in a potential misclassification between preterm and full term delivery), a paediatrician, who will be blinded to exposure will be asked to assess the pregnancy dating

data and where possible examine the infant in order to determine the gestational age of the infant at birth (within 24-48 hours of birth).

11.3.8 Congenital Anomaly

The frequency of major congenital anomaly, defined as a structural abnormality present at birth, was reported at 2% in a Ugandan cohort (Ndibazza, Lule *et al*. 2011). Whilst DolPHIN-2 is not designed to capture major congenital anomalies associated with DTG (by study design since the fetus is fully formed at the time or recruitment in the third trimester, and by sample size which is not powered to detect anomalies occurring at low frequency in the population), we will assess all reported anomalies. Where necessary, and if the woman provides consent, photographs of the congenital anomaly/ies will be reviewed by an independent review panel including a paediatrician and a geneticist. All congenital anomalies for DTG exposed infants should reported to the Antiretroviral Pregnancy Registry (APR) (See Section 11.4.1).

11.3.9 Neonatal death

In the event of neonatal death, if the mother consents, a verbal autopsy will be performed by the study team in attempt to elucidate the cause of death.

11.3.10 IRIS

As case definitions for Immune Reconstitution Inflammatory Syndrome (IRIS) vary and rely heavily on clinical judgement, we will report all AEs requiring hospitalisation (or of greater severity) to the IDSMB as per standard practice. Based on IDSMB review, we will assess the incidence of IRIS retrospectively.

11.4 Safety Reporting

11.4.1 Pregnancy Reporting

Pregnancy complications (e.g. preeclampsia or eclampsia, prolonged hospitalization after delivery, for wound infections etc, seizures) and elective terminations for medical reasons must be reported as an AE (Section 11.1) or SAE (Section 11.2). Spontaneous abortions and stillbirths must be reported as an SAE (Section 11.2).

Additionally, investigators are encouraged to register each maternal participant with the APR prospectively as soon as possible after the participant starts treatment as part of the study on Day 1, and before the pregnancy outcome (otherwise there is less likelihood of the participant being included in the APR). More information including copies of applicable CRFs and fax numbers are available at www.apregistry.com.

Participants who develop a subsequent pregnancy will transition to a nationally recommended ART regimen and will remain in the study for safety follow-up. If DTG or EFV exposure has occurred, the pregnancy will be recorded with the APR and followed to ascertain outcomes. As above, spontaneous abortions and stillbirths relating to subsequent pregnancies will be reported as SAEs.

11.5 Adverse Event Reporting

All clinically significant observed or volunteered AEs regardless of treatment group or suspected causal relationship will be reported. For all AEs, the investigator must pursue and obtain information adequate both to determine the outcome of the AE and to assess whether it meets the criteria for classification as an SAE requiring immediate notification.

Each AE is to be assessed to determine if it meets the criteria for SAEs. If an SAE occurs that meets the IRB, local and international regulations for reporting, expedited reporting will be done. All SAEs will be reported to the sponsor within 24 hours of knowing about the event. Further relevant follow-up information will be given as soon as possible. Follow-up will continue until the event resolves. AEs will be followed up for 30 days and SAEs will be followed up for 6 weeks following the last prescribed dose of the study unless resolved.

All SAEs must be reported immediately. The SAEs should be reported immediately to the Principal Investigator (within 24 hours of a member of the study team becoming aware of the event). A SAE form should be completed, and an assessment of whether the SAE is a Suspected Unexpected Serious Adverse Reaction (SUSAR) conducted. The Principal Investigator is responsible for determining whether the SAE is a Suspected Unexpected Serious Adverse Reaction (SUSAR) and for reporting this in accordance with Good Clinical Practice (GCP) and applicable regulatory requirements. All SAEs must be reported in accordance with local protocols for reporting adverse study events.

All AEs will be reported on the AE page(s) of the CRF. For example, the same AE term should be used on both forms. AEs should be reported using concise medical terminology on the CRFs as well as on the form for collection of SAE information.

Pre-term deliveries at less than 34 weeks as determined by the Ballard score will be reported as SAEs.

11.6 Management of Post-recruitment Illness

All participants with post-recruitment illness will be monitored until symptoms resolve, laboratory changes return to baseline or until there is a satisfactory explanation for the changes observed. Participants will receive medical care including admission at the hospital associated with the study and participants will be managed in accordance with national treatment guidelines.

11.7 TRIO EXTENSION MANAGEMENT OF ADVERSE EVENTS

Adverse and Serious Adverse events should be managed as per section 11.1 to 11.6 of the DolPHIN 2 protocol.

12.SUBSTUDIES

Participants in DolPHIN-2 will be invited to participate in sub-studies exploring aspects of access to and response to treatment.

12.1 Pharmacokinetic and Pharmacogenomic Substudy

Storage of maternal plasma and breast milk from all participants will enable the undertaking of substudies to investigate reasons for poor treatment response or development of toxicities.

Maternal plasma will be stored at each time point. Among women who have a poor response to treatment, either in terms of viral kinetics, transmission of HIV to their infant, or toxicities, a pharmacokinetic analysis will be undertaken to determine whether these responses can be explained in terms of drug exposure. Women with these poor outcomes will be matched (by age and gestation of presentation) with mothers who responded well to DTG without any adverse events.

Similarly, breast milk will have been stored to enable quantitation of drug in this compartment. Breast milk cells will be stored as a pellet to enable analysis of cell-associated DNA in addition to the cell-free RNA in relation to drug penetration.

Our sample collection strategy is designed to be pragmatic and convenient: sparse PK sampling (with dosing history) will coincide with safety bloods, and will be collected in all mothers and stored. Cervicovaginal fluid will be sampled antenatally. Samples will be retrieved in mother-infant pairs where transmission, toxicity or adverse outcomes have been documented (alongside matched controls). In addition, a subset of samples will be analysed using population PK models to i) evaluate for likely non-adherence, and ii) to assess covariates of drug exposure. Paired maternal plasma: cord blood will be collected at delivery whenever possible at the Cape Town site, and stored.

Infant bloods: safety bloods are being collected at 6 weeks postpartum. Dried blood spots and plasma will be stored.

We will develop population PK models for DTG in pregnancy; rich PK data from DolPHIN-1 will be used to develop a structural model for DTG (NONMEM). Sparse data will be fitted to determine covariates (eg maternal weight, renal and liver function, HBV coinfection) and also to assess for likely non-adherence. In addition we have developed a PBPK model of maternal-to-milk-to-infant transfer of EFV (Olagunju, Siccardi *et al*. 2014); similar work will be undertaken for DTG. Compartmental PK (milk, cord blood) modelling will also be integrated, using DolPHIN-2 samples (we will analyse unbound fraction of DTG in cord blood in a subset of infants)

PD outcomes of interest are: adverse maternal or infant drug reactions, MTCT, lack of VL suppression at delivery and 6w post-partum, detectable viraemia in milk (or milk: plasma VL discordance). PK parameters used for PK-PD are: Bayesian estimates of CL/F for relationship

with VL, and absolute concentrations in infants/milk for toxicities, and transmissions (where endpoints are anticipated to be few).

Pharmacogenetic study: Maternal blood will be stored for analysis of polymorphisms which may account for differences in treatment response and toxicity development.

12.2 Qualitative Analysis of Factors Associated with Late Presentation

The effect of late presentation on future adherence is of particular concern. Predictors for disengagement postpartum were studied by Phillips *et al*, 2014, in South Africa and reported a 4% increased hazard (HR: 1.04 CI: 1.00-1.07) for every additional increase in gestational age week at ART initiation. This research suggests that presentation in late pregnancy may indicate wider issues in health-seeking behaviour, and may highlight a high-risk population that needs focused strategies to improve adherence postpartum.

However, more in-depth qualitative studies are needed to broaden our knowledge of these barriers to early antenatal presentation and the effects on future adherence, from both the perspective of pregnant women and healthcare workers, and how to combat them. This work aims to identify challenges and potential solutions to the implementation of evidence and recommendations arising from the clinical trial.

The overall aim of this work is to inform programmatic guidance and policymakers in ensuring equity, uptake and sustainability of services.

Selected participant from DolPHIN-2 will be invited to participate in more detailed interviews, and separate consent will be sought from them at this point.

Quantitative research methods will evaluate equity of access, and whether late presenting pregnant women are less likely to engage with and adhere to cART. Community-based studies will identify patient characteristics that predict poor adherence and retention – including remote location, lack of partner disclosure, domestic violence, youth, migration for work, substance use. The aim of this work is to understand barriers and priorities and what counselling messages/language and resources are needed when trying to support these women. We will extend these studies to include partners of pregnant women, in order to understand the factors associated with horizontal, and vertical transmission in this group of individuals, and whether risk compensation occurs when using a potent drug such as DTG. Within clinical trial participant, we will also assess if the main driver of outcome is rate of suppression or rate of adherence because of tolerability.

Qualitative studies will investigate the acceptability and feasibility of DTG in late pregnancy. Qualitative research methods provide useful insights for programmers and policymakers including an ability to assess barriers and enablers to the uptake of a new drug regimen. These methods are able to explore research participants' lived realities and allow researchers to understand how social experience is created and understood in everyday life, including voices of service users and communities. We will conduct formative research in the two country

contexts to explore the perceptions and experiences of pregnant women, community members and health workers on the factors associated with late presentation in pregnancy and adherence to ART. We will also interview formal health care workers (primarily midwives, pharmacists and supervisors) and key informants (Ministry of Health, donors, NGOs and other stakeholders) to explore in more depth perceptions of the feasibility and potential impact of DTG in late pregnancy; the processes required before practice can change in a country and the key enablers and barriers to change. The overall aim of this work is to inform programmatic guidance and policymakers in ensuring equity, uptake and sustainability of services.

The qualitative work involving participants who are not included in the DolPHIN-2 clinical trial (healthcare providers, key informants etc.) will undergo separate ethical approval.

12.3 Genital fluid Substudy

We will sample cervicovaginal fluid from as many as possible women enrolled into DolPHIN-2, to evaluate:

- HIV viral dynamics (viral load, genetic and phylogenetic characterisation, viral phenotype),
- Pharmacokinetics
- Vaginal microbiome, and its relationship to viral shedding and pre-term labour

Consent will be sought from women enrolled in DolPHIN-2 to provide samples of cervicovaginal fluid. Given this is a secondary endpoint, lack of willingness to participate in this aspect of the study does not preclude enrolment in DolPHIN-2. We will sample from all the participants who provide consent. This sample size is ample for this type of exploratory virological research.

Participants will be asked for separate consent to sample cervicovaginal fluid (up to 2.5 mLs) at screening and at 4 weeks. Samples will be analysed with maternal plasma already collected at these timepoints in the main study. This will enable characterisation of the female genital tract as a viral/ drug compartment.

Women who have given cervicovaginal fluid samples will be approached to give consent for an infant throat and stool swab. The additional infant samples will be analysed to assess the influence of ART on infant gut flora.

12.4 Ultrasonography Substudy

Women will be offered repeat ultrasonography at one and four weeks after commencement of ART. This will enable assessment of fetal growth, placental blood flow and other variables of late pregnancy which may be influenced by pharmacotherapy.

12.5 Cord Blood and Placenta Substudy

Where possible, paired maternal and cord blood, together with a sample of placental tissue will be collected at delivery. The placenta plays a major role in protecting the foetus from maternal

rejection and from infections, including HIV (Suchard, Mayne *et al*. 2010). One of the main roles of the placenta is to maintain fetal-maternal (FM) tolerance and it is speculated that HIV/ART interferes with FM tolerance at the placental interface. Immunological assessments will be carried out on the collected placenta to investigate any disruption to immune regulation mechanisms.

12.6 Neuro-imaging Substudy

HIV-associated functional and structural changes in the central nervous system are well-documented. In addition, EFV is associated with specific neuropsychiatric side effects, particularly early in the course of treatment, and DTG is increasingly recognised to be associated with different neurological side effects. To compare the effects of DTG and EFV, we are investigating the comparison of DTG and EFV in a subsample of postpartum women enrolled in to the main trial. Women will be assessed using (a) formal neuropsychiatric testing as well as neuroimaging. A separate protocol will be developed for this substudy.

12.7 Health Economic Analysis

To understand the cost-effectiveness of DTG in this population, data from the DolPHIN-2 study will be used as inputs for the health economic analysis. To capture quality of life information, quality of life questionnaires will be administered that include the validated Medical Outcome Study – HIV Health Survey (MOS-HIV, Appendix 8) at four weeks following commencement of ART and after 24 and 48 weeks postpartum. Social economic data will be captured using a questionnaire derived from the DHS survey. Utilisation costs will be obtained via the recording of adverse events, including consultations, hospitalisations, investigations, procedures and prescriptions; these data will be captured routinely throughout the DolPHIN-2 study.

Using decision-based analytical models and a national health systems perspective, we propose to evaluate:

(i) the cost-effectiveness of life-long DTG vs EFV-based cART
(ii) the cost-effectiveness of increasing access to DTG-based cART for PMTCT among women without access due to lack of drugs, lack of local health-care centres or lack of knowledge of HIV infection.

Overhead costs to overcome barriers to access to PMTCT will be obtained from previously published analyses. Clinical inputs (probability of MTCT) will be obtained from DolPHIN-2. Estimates of the disability-adjusted life years (DALYs) associated with MTCT will be based on the HIV-attributable reduction in life expectancy plus standard disability weights for HIV infection and AIDS. Based on the differences in the probability of MTCT with each treatment alternative, we will calculate the treatment-related costs offsets arising from the need to provide health care services to fewer HIV positive infants in the future. For example, for each child requiring ART in Uganda, the discounted lifetime cost of ART is ~$3800, with a mean life expectancy of around 15 years. Based on the discounted differences in life expectancy between HIV-negative children and HIV-positive children who are left untreated, or given ART, a child infection can be estimated to be associated with 23.70 DALYs. Cost effectiveness analysis will yield the incremental costs per DALY averted using DTG-based ART versus standard of care.

12.8 Prescribing Support

Prescribing Support will aid the deployment of DTG in Africa. Alongside the clinical trial, we will gauge the scope, and level of support required, and evaluate whether existing mechanisms are able to deliver the skills and knowledge required. This includes management of toxicities and drug interactions. We will evaluate drug interactions education within training packs for national programmes, and explore how the Liverpool HIV Drug Interaction resource (www.hiv-druginteractions.org) can be more widely deployed into rural prescribing communities though phone Apps, integration into existing systems in clinics and pharmacies, by lay workers in the community and by people with HIV themselves.

12.9 Contraception Substudy

There are significant drug drug interactions (DDIs) with ARVs and hormonal contraceptives. In contrast to combined oral contraception, DDIs with depomedroxyprogesterone (DMPA) and implants (etonogestrel [ENG] and levonorgestrel [LNG]) are poorly studied. Hormonal contraception (particularly injectable, or implant) is routinely offered following infant delivery (and in South Africa often during the 3rd stage of labour) as part of a holistic approach to public health and family planning. As such, contraception choices, safety and efficacy lies within the broader evaluation of the interventions being assessed in DolPHIN-2.

This substudy aims to collect data from women choosing the hormonal contraceptives DMPA and LNG to elicit the pharmacokinetics of the ARVs and contraceptives.

A separate protocol will be developed for intensive pharmacokinetic sampling in this substudy. Participants will be asked to consent to an additional blood sample for sparse pharmacokinetic sampling at postpartum visits if they have chosen the hormonal contraception DMPA or LNG.

12.10 TRIO EXTENSION SUBSTUDIES

There are no sub studies for the Dolphin 2 TRIO extension

13.DATA AND STATISTICAL ANALYSIS

Statistical analysis will be done by the trial statistician at tCTU. A general description of the statistical methods to be used to analyze the efficacy and safety data is outlined below. Specific details will be provided in the Statistical Analysis Plan (SAP). Statistical analysis of each substudy will be described in detail separately in its own SAP.

The primary study objective will be addressed by comparing the proportion of participants having a primary efficacy endpoint between treatment groups. Specifically, the following primary statistical hypotheses will be tested.

Null hypothesis H_0: there is no difference between treatments in the proportion of participants having a primary efficacy endpoint.

Alternative hypothesis H_a: The proportions of participants having a primary efficacy endpoint are different between the two treatment groups.

Analysis methods and decision rules are outlined in Section 13.3 below.

13.1Analysis Populations

Participant Population
Intention-to-treat (ITT) population: This participant population consists of all consented eligible participants (not including those who are withdrawn at visit 2 due to ineligibility)

Per-protocol (PP) population: this population is a subset of the ITT population. Participants with major protocol deviations will be excluded from PP population. Major protocol deviations will be defined in the SAP.

Safety population: this population consists of all randomized participants who receive at least one dose or partial dose of study drug.

13.2 Sample Size Determination

Sample size calculations are detailed in Section 6.1.

13.3 Efficacy Analyses

13.3.1 Primary Endpoint

Determination of the primary endpoint will be based on the FDA 'snapshot' algorithm (modified to include sensitivity analysis for duration of therapy prior to delivery. The primary endpoint will be summarised by number (%) of participants with events and compared by a log binomial model (a generalised linear model [GLM]) that includes treatment as a study variable, VL (≥ or <median copies) and (CD4 ≥ or <200 cells/mm3) as covariates, which will generate risk ratio (RR) together with their 95%CIs of having a primary outcome between DTG and EFV groups.

Homogeneity of treatment effects, both in RR and direction, in the following subgroups will be assessed by subgroup analysis. Analysis methods will be detailed in the SAP.

- Age (< or ≥ median in years)
- Country (South Africa vs Uganda)
- VL (≥ or < 100,000 copies)
- CD4 (≥ or <200 cells/mm3)
- Gestational age (< or ≥ median)

In addition, covariate adjustment will be performed by GLM model with treatment as a study

variable and covariates listed above (with the exception of VL) as covariates. From this model, adjusted RR together with their 95% confidence interval will be derived.

In addition, the risk difference will be estimated using GLM model with binomial distribution and identity link function. The GLM model will have treatment as a study variable, VL (≥ or <median copies) and (CD4 ≥ or <200 cells/mm3) as covariates, from which risk difference and its 95% confidence interval will be derived.

The primary endpoint analysis will be based on ITT population. An additional analysis of the primary outcome will also be presented based on the per-protocol population. A number of sensitivity analyses will be conducted to assess the robustness of the primary efficacy analysis. These include a crude GLM model analysis with only treatment as a predictor and the GLM model on the per-protocol participant population. Additional post-hoc analyses may be conducted to investigate unexpected results.

13.3.2 Secondary Endpoints

The secondary outcomes will be analysed similarly as the primary endpoint analysis using a GLM. For GLM analysis of a continuous endpoint such as quality of life, normal distribution and identity link functions will be used; For GEE analysis of a binary outcome (such as having a primary endpoint), binomial distribution and log link functions will be used.

For the analysis of time-to-event outcome, the Kaplan-Meier curves will be presented and compared by the log rank test by treatment group and hazard ratio and its 95% confidence interval will be calculated using Cox regression model with the treatment arm as the study variable and VL≥ or <median, and CD4 ≥ or <200 cells/mm3 as covariates.

The secondary binary outcomes with repeated measurements such as virological responses at different time points will be summarised using number (%) of events at each time point and analysed using a generalised estimating equation (GEE) model, in which treatment, time, interaction between treatment and time as fixed effects and participant as random effect. Exchangeable covariance structure will be used. The odds ratio together with their 95%CIs at each time point will be derived.

Continuous variables will be summarised using number of observations, mean (standard deviation), median (IQR); categorical variables will be summarised by the number and percentage of events. Time-to-event variables will also be summarised by the number (%) of participants having an event and events per 100 person-years by treatment arm.

13.4 Safety Analyses

13.4.1 Primary Safety Endpoint

The primary safety endpoint will be summarised by number (%) of participants with events and compared by a GLM with binomial distribution and identity link function. The GLM model will have treatment as a study variable, VL (≥ or <median copies) and (CD4 ≥ or <200 cells/mm3) as covariates, from which risk difference between DTG and EFV groups and its

95% confidence interval will be derived.

13.4.2 Adverse Events

Because the safety profile of the study drug has been well established in previous large and extensive trials, this study will collect limited adverse event data. For adverse events that are collected as specified in Section 11.1 Adverse Events, the verbatim terms reported in the CRF by investigators to identify adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA). For each MedDRA preferred term, the percentage of participants who report at least 1 occurrence of the given event will be summarized by treatment group. Additional summaries, listings, or participant narratives may be provided, as appropriate.

13.4.3 Clinical Laboratory Tests

The individual laboratory findings and change from baseline will be summarized descriptively by treatment. For the repeated values, the first observed value will be used for generating summary statistics

13.5 Independent Data Safety Monitoring Board (IDSMB)

An IDSMB will be established to monitor data on an ongoing basis to ensure the continuing safety of mothers and children enrolled in this study. The IDSMB will review unblinded safety data periodically. If necessary or requested by the IDSMB, mother and child level unblinded data may be provided to the IDSMB

The membership of the IDSMB is limited and includes an independent Chair (not involved directly in the trial other than as a member of the IDSMB), at least one medical expert in the relevant therapeutic area and at least one independent statistician. The Organogram in Section 14 summarises the function and accountability between the IDSMB, the TSC and TMG and all other vital components of the DolPHIN-2 trial.

In their first meeting the committee will decide the frequency of future meetings, the scope of safety review, and finalize the charter. Detailed IDSMB responsibilities, authorities, decision guidelines, and procedures will be documented in its charter. These include: review of trial protocol; review of all relevant ethical issues relating to the trial; review of study recruitment and losses to follow-up; assessment of quality of study execution, including completeness of data; monitor compliance with the protocol by participant and investigators; monitor (together with the TSC) evidence for treatment differences in main efficacy outcome measures; monitor (together with the TSC) for treatment harm (toxicity data); decide whether to recommend that the trial continues, stops or is modified; suggest additional data analyses; advise on further protocol modifications; monitor planned sample size assumptions; monitor compliance with previous IDSMB recommendations; assess impact and relevance of external evidence. At each meeting the IDSMB will review laboratory and clinical data relating to mother and child. Further IDSMB meetings may be convened (at the discretion of the IDSMB chair, or upon request from the Trial Steering Committee) if unexpected data emerge, or following any modification of the study. Any Serious Adverse Events (SAEs) or Suspected Unexpected

Serious Adverse Reaction (SUSARs) (see Section 11) will be reported to the IDSMB in a timely manner.

The IDSMB will be supported by an independent statistician which is not involved with the study otherwise. All unblinded data will be handled by this statistician until the completion of the study.

13.6 Interim Analysis

The IDSMB will undertake a single planned interim (after 125 women have data available on the primary endpoint), using the alpha spending function to preserve the overall two-sided type I error rate (α=0.05) for effectiveness. The α spent in the interim analysis is 0.001 with a corresponding critical value of 3.2905 for Z-test. If significance is found in favour of active treatment, the study may be recommended for stopping with sufficient statistical evidence of efficacy. If the 0.001 level of significance is not reached at the interim, the study continues beyond the first interim look to normal completion, at which time the hypothesis test is conducted at significance level of 0.0498 (the critical value for the final analysis is 1.9619).

All SAEs and SUSARs (including any congenital anomalies and other serious adverse infant outcomes) will be reported to the Sponsor within 24h, and will also be reported in a timely manner to the IDSMB. The IDSMB may trigger further safety reviews as it sees fit, taking into account accrued safety data from this study, as well as the sum of current knowledge around the safety of dolutegravir in pregnancy.

13.7 TRIO EXTENSION DATA AND STATISTICAL ANALYSIS

Datasets from the ADVANCE, NAMSAL and DolPHIN-2 trials will be combined every 6 months, from the start of the study.

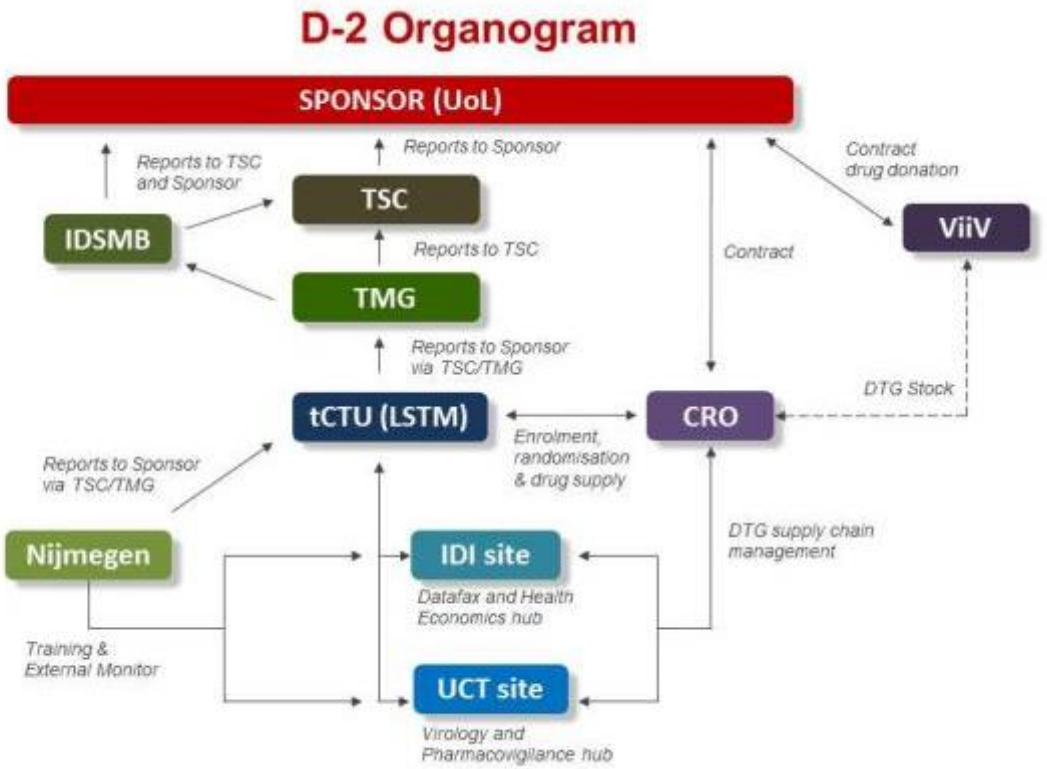
At each stage, the key safety endpoints will be compared between people taking DTG or EFV, and between TAF or TDF where appropriate. The number of participants reporting key clinical adverse events (e.g. diabetes, myocardial infarction, birth defects, fractures) will be compared between the arms of each trial, and in the combined analysis.

Adverse birth outcomes (maternal and infant) will be summarised by treatment arm and trial, and combined in a meta-analysis of the three trials. HIV RNA suppression and the incidence of Mother to Child Transmission of HIV (MTCT) will also be summarised by treatment arm and trial. This analysis will include HIV infections occurring post-partum, during breastfeeding.

Changes in laboratory and other markers (e.g. lipids, glucose,) will also be compared between TAF-LD, TLD and TLE. ADVANCE is the only trial evaluating TAF-LD, and the planned mass rollout of TAF in Zambia and Botswana makes evaluation of this combination a priority.

14.STUDY MANAGEMENT

The organogram summarises the DolPHIN-2 management structures



The day-to-day management of the DolPHIN-2 study will be undertaken by the Study Team, comprising the Chief investigator, all co-investigators, the tCTU at Liverpool School of Tropical Medicine, and expert pharmacists involved in safety monitoring. The Project Manager will meet with the tCTU fortnightly (face to face or teleconference) to cross-check all documentation regarding screening, randomisation and enrolment.

The study site will be responsible for conducting the trial. The study site will collaborate with the affiliated antenatal clinics for this project for patient recruitment and follow up. Safety laboratory tests will be performed at laboratories which have achieved appropriate accreditation by the relevant professional body.

14.1 Trial Steering Committee (TSC)

A Trial Steering Committee (TSC) with an independent Chair will oversee the conduct of the study, and will receive reports from the Study Team, and the IDSMB. The TSC is the oversight body for the trial, whereas the TMG is the body responsible for operationalising and executing the trial. The TSC, acting on its own judgement or upon any recommendation from the IDSMB, will be able to recommend to the Sponsor that the study is continued, stopped or amended. The TSC has the following responsibilities: review and approve trial protocol; review

all relevant ethical issues relating to the trial; review progress of study recruitment and losses to follow-up; assess quality of study execution, including completeness of data; monitor compliance with the protocol by participant; consider recommendations from the IDSMB; advise the TMG on all aspects of the trial; decide whether to recommend that the trial continues, stops or is modified; suggest additional data analyses; advise on further protocol modifications; monitor planned sample size assumptions; monitor compliance with previous IDSMB recommendations; assess impact and relevance of external evidence; review and approve all substudies; review and approve all study-related outputs (including substudies) for submission to conferences, as abstracts or presentations or written manuscripts and briefing documents.

The TSC Terms of Reference will outline responsibilities and duties as well as confidentiality and data protection. The TSC will be chaired by an independent expert with relevant experience and no other direct responsibility for the DolPHIN-2 trial, and will contain independent experts, a community member, the chief investigator as well as the South African and Ugandan PI (or if need be, will deputise to the local co-I).

14.2 Sample Processing

Material transfer agreements will underpin any shipment of samples. The current version of the DolPHIN-2 trial Laboratory Handbook will provide full details of the steps involved in collection, processing, shipment, storage, receipt, analysis, and reporting of all samples.

14.3 Clinical Monitoring

At each study day visit including screening, the patient will be assessed by the research physician or nurse. In addition, participant will be given details of how to reach a member of the study team in an emergency.

14.4 Laboratory Monitoring

Safety bloods (haemoglobin, white cell count, platelets, creatinine, bilirubin, ALT, CK) will be performed regularly. Additionally, should an adverse event be reported, further samples will be taken as soon as practically possible. The anti-retroviral efficacy of the novel combination compared with standard of care will be monitored by measurement of HIV viral load at baseline, and delivery. Severity of abnormalities will be defined according to the DAIDS criteria in Appendix 5.

14.5 Termination of the trial

The Sponsor may terminate either part of, or the entire study for safety or administrative reasons. A written statement fully documenting the reasons for such a termination will be provided to the Ethics Committee and the Regulatory Authority as appropriate.

14.6 TRIO EXTENSION STUDY MANAGEMENT

Study Management is as per Section 14.1 to 14.5 of the main DolPHIN 2 Protocol

15.DATA HANDLING

15.1 Recording of Data

The data collected during the study will be recorded in an individual participant specific Case Report Form (CRF). In order to maintain confidentiality, the participant will be identified only by participant number and initials on the CRF. All CRFs should be completed legibly by an appropriate member of the study team who should be identified. A CRF must be completed for each participant who signs a consent form and undergoes any screening procedures.

Corrections to the data on the CRF will only be made by drawing a single line through the incorrect data (so as not to obscure the original entry) and inserting the correct data next to the original entry. The incorrect data must never be obliterated using correction fluid. Each correction will be initialled and dated by the person making the correction. CRFs will be the responsibility of the study teams at each site; in both Uganda and South Africa, the study co-ordinator will manage the study team on a day-to-day basis, overseen by the local PI (Lamorde and Myer, respectively).

15.2 Source Documentation and Study Records

The participant's number and date of entry into the study, along with a study identifier, should be recorded in the participant's study records. The following should also be recorded in the study records; confirmation of written consent, the participant's clinical status, date of every study visit, date study medication was given, concomitant medications, copies of all relevant reports and laboratory tests, comments on results and reference to any adverse events. Data will be partly collected as source data on the CRF, it will be clearly documented in the Investigator Site File which data will be collected as source data only.

15.3 Data management

The CRFs developed for the DolPHIN-2 study are designed for use with the DataFax data management system. Datafax forms will be transmitted to the DataFax unit at IDI and study data will be entered into a study specific database by designated staff on a regular basis from completed CRFs, to ensure that it is up to date. The database will be kept on a secure PC. Access to the database will be given to authorised personnel only and a log of authorised personnel will be stored in the trial master file. Any data anomalies or values found to be outside normal ranges will be checked with the investigator. When corrections are required, they will be made on CRF and the study database will be amended.

Data sharing with other studies is allowed, on discretion of the study team, who will evaluate potential collaborations on a case by case basis, depending on the scientific merit, research governance, costs and data quality. The guiding principle will always be to maximize the wider benefits of this study wherever possible.

15.4 Archiving and storage of data

Following completion of the study, participant records, CRF, and other study documentation will be retained by the investigator in accordance with Good Clinical Practice and applicable regulatory requirements.

15.5 TRIO EXTENSION DATA HANDLING

Data Handling is as per Section 15.1 to 15.4 of the main DolPHIN 2 Protocol

16. QUALITY CONTROL AND QUALITY ASSURANCE

16.1 Monitoring Arrangements

The purpose of monitoring is to verify that the rights and well-being of human participants are protected; that trial data is accurate, complete and verifiable with source data; that the trial is conducted in compliance with the protocol, GCP, and the applicable regulatory requirements. The Sponsor will act as monitor of the study. The Investigator must agree to allow the study monitor to inspect all CRF and corresponding source documents, e.g. original medical records, participant records and laboratory raw data, access to the clinical supplies, dispensing and storage areas and agree to assist with their activities if requested. The investigator should provide adequate time and space for monitoring visits.

One external and one or more internal monitors per site will be assigned to the study. The external monitor, as a representative of the Sponsor, has the obligation to follow the study closely. The monitor will visit the site at regular intervals and will be in contact by phone and written communication, as required.

Site investigators and designated study personnel will allow the study monitors to inspect study documents, pertinent hospital or clinic records as well as site facilities, as required. All aspects of the study will be carefully monitored in order to ensure compliance with Good Clinical Practice and all applicable regulatory guidelines. The monitor will be responsible for verification of:

- adequacy of study personnel's qualifications as well as facilities,
- informed consent procedures and patient eligibility
- the accuracy and completeness of the CRF entries, source documents and other trial-related records against each other,
- appropriate IMP storage, usage and accountability,
- appropriate adverse event reporting
- maintenance of the essential documents
- all other aspects of the trial relating to protection of the rights and well-being of participant, accuracy of trial data and adherence to the protocol, GCP and applicable regulatory requirements

The monitoring plan will define the percentage of verification; however, informed consents and patient eligibility will be checked 100%. The study database will only be locked after the data have been monitored by the sponsor and all queries issued through data cleaning activities have been completed and resolutions documented.

16.2 Quality Assurance

For the purpose of compliance with GCP and regulatory agency guidelines, it may be necessary for sponsor authorised Quality Assurance personnel and/or authorised personnel from an external regulatory agency to conduct an audit/inspection of an investigational site. The purpose of an audit is to assess the quality of data with regard to accuracy, adequacy and consistency, and to assure that studies are in accordance with GCP and Regulatory Agency guidelines. Having the highest quality data from studies is an essential aspect of drug development.

The Investigator will be given sufficient notice to prepare for such visits, which are planned to take usually between one and two days and may be conducted at any stage during the study. The audit will involve the review of all study related documentation, which is required by GCP to be maintained by each site, review of drug storage, dispensing and return, review of all study related supplies and review of source documents against the CRF to assure the adequacy and accuracy of the information which has been recorded, including the verification of any AE which have occurred.

16.3 TRIO EXTENSION QUALITY CONTROL & QUALITY ASSURANCE

Quality Assurance is as per Section 16.1 to 16.2 of the main DolPHIN 2 Protocol

17.ETHICS & ADMINISTRATION

The study protocol, participant information and consent form, available safety information, participant recruitment procedures (e.g. advertisements), information about reimbursement and compensation available to the participant and documentation evidencing the investigator's qualifications will be submitted to the Ethics Committees for ethics review and approval according to local regulations, prior to the study start. Any changes, which may need to be made, will be submitted in the form of numbered and dated protocol amendments in accordance with local regulations.

17.1 TRIO EXTENSION ETHICS & ADMINISTRATION

Ethical Approval is as per Section 17 of the main DolPHIN 2 Protocol

18.REGULATORY APPROVAL

As required by local regulations, approval of the appropriate regulatory bodies will be obtained, prior to study initiation.

18.1 Publication Policy and Dissemination Plan

The study has developed a communication plan based on a stakeholder analysis conducted in the study development phase. Information will be developed in modalities appropriate to the target audience. At both study sites, Community Advisory Boards (CAB) have been involved during the study planning, and will continue to provide input at all phases of the study from set-up through to dissemination. Through regular meetings with the study team, key updates on study progress, new evidence external to the study, and study results will be communicated and presented in a format which is accessible to members of the community.

Other activities supporting communication regarding the DolPHIN-2 study will be headed by the Liverpool team and will include the DolPHIN-2 website including the project blog, Twitter and Facebook pages, a project brochure, a project newsletter produced quarterly and distributed via the website, social media and to key stakeholders and press releases to be produced regularly when there is a significant project update such as a key finding.

A whole or part of this study results will be communicated, orally presented, and/or published in appropriate scientific journals. Full anonymity of participant's details will be maintained throughout. All communications will be available on our website www.dolphin2.org.

18.2 Drug Accountability

The investigator will ensure that the IP will only be used in accordance with the protocol. Drug supplies will be kept in a secure, limited access storage area under the recommended storage conditions. A drug accountability log will be kept with the investigational supplies for reconciliation purposes. This should be used to record the identification of the participant to whom the IP was dispensed, the date and quantity of IP dispensed. This will be verified by the study monitor.

18.3 Financial Aspects

This trial is being funded by UNITAID. The University of Liverpool will administer the funds, act as Sponsor for this study, and provide appropriate insurance in accordance with current arrangements for clinical trials conducted in sub-Saharan Africa.

18.1 TRIO EXTENSION REGULATORY APPROVAL

Regulatory Approval is as per Section 18.1 to 18.3 of the main DolPHIN 2 Protocol

19.REFERENCES

Allen, E. N., C. I. Chandler, N. Mandimika, C. Pace, U. Mehta and K. I. Barnes (2013). "Evaluating harm associated with anti-malarial drugs: a survey of methods used by clinical researchers to elicit, assess and record participant-reported adverse events and related data." Malar J **12**: 325.

Allen, E. N., M. Gomes, L. Yevoo, O. Egesah, C. Clerk, J. Byamugisha, A. Mbonye, E. Were, U. Mehta and L. M. Atuyambe (2014). "Influences on participant reporting in the World Health Organisation drugs exposure pregnancy registry; a qualitative study." BMC Health Serv Res **14**: 525.

Cahn, P., A. L. Pozniak, H. Mingrone, A. Shuldyakov, C. Brites, J. F. Andrade-Villanueva, G. Richmond, C. B. Buendia, J. Fourie, M. Ramgopal, D. Hagins, F. Felizarta, J. Madruga, T. Reuter, T. Newman, C. B. Small, J. Lombaard, B. Grinsztejn, D. Dorey, M. Underwood, S. Griffith, S. Min and S. S. T. extended (2013). "Dolutegravir versus raltegravir in antiretroviral-experienced, integrase-inhibitor-naive adults with HIV: week 48 results from the randomised, double-blind, non-inferiority SAILING study." Lancet **382**(9893): 700-708.

Castagna, A., F. Maggiolo, G. Penco, D. Wright, A. Mills, R. Grossberg, J. M. Molina, J. Chas, J. Durant, S. Moreno, M. Doroana, M. Ait-Khaled, J. Huang, S. Min, I. Song, C. Vavro, G. Nichols, J. M. Yeo and V.-S. Group (2014). "Dolutegravir in antiretroviral-experienced patients with raltegravir- and/or elvitegravir-resistant HIV-1: 24-week results of the phase III VIKING-3 study." J Infect Dis **210**(3): 354-362.

Castellino, S., L. Moss, D. Wagner, J. Borland, I. Song, S. Chen, Y. Lou, S. S. Min, I. Goljer, A. Culp, S. C. Piscitelli and P. M. Savina (2013). "Metabolism, Excretion, and Mass Balance of the HIV-1 Integrase Inhibitor, Dolutegravir, in Humans." Antimicrob Agents Chemother.

Chan, A. K., E. Kanike, R. Bedell, I. Mayuni, R. Manyera, W. Mlotha, A. D. Harries, J. J. van Oosterhout and M. van Lettow (2016). "Same day HIV diagnosis and antiretroviral therapy initiation affects retention in Option B+ prevention of mother-to-child transmission services at antenatal care in Zomba District, Malawi." J Int AIDS Soc **19**(1): 20672.

de Boer, M. G., G. E. van den Berk, N. van Holten, J. E. Oryszcyn, W. Dorama, D. A. Moha and K. Brinkman (2016). "Intolerance of dolutegravir-containing combination antiretroviral therapy regimens in real-life clinical practice." AIDS **30**(18): 2831-2834.

de Ruiter, A., G. P. Taylor, P. Clayden, J. Dhar, K. Gandhi, Y. Gilleece, K. Harding, P. Hay, J. Kennedy, N. Low-Beer, H. Lyall, A. Palfreeman, S. O'Shea, P. Tookey, J. Tosswill, S. Welch, E. Wilkins and H. I. V. A. British (2014). "British HIV Association guidelines for the management of HIV infection in pregnant women 2012 (2014 interim review)." HIV Med **15 Suppl 4**: 1-77.

Dooley, K. E., P. Sayre, J. Borland, E. Purdy, S. Chen, I. Song, A. Peppercorn, S. Everts, S. Piscitelli and C. Flexner (2013). "Safety, tolerability, and pharmacokinetics of the HIV integrase inhibitor dolutegravir given twice daily with rifampin or once daily with rifabutin: results of a phase 1 study among healthy subjects." J Acquir Immune Defic Syndr **62**(1): 21-27.

Ford, N., L. Mofenson, K. Kranzer, L. Medu, L. Frigati, E. J. Mills and A. Calmy (2010). "Safety of efavirenz in first-trimester of pregnancy: a systematic review and meta-analysis of outcomes from observational cohorts." AIDS **24**(10): 1461-1470.

Garcia, P. M., L. A. Kalish, J. Pitt, H. Minkoff, T. C. Quinn, S. K. Burchett, J. Kornegay, B. Jackson, J. Moye, C. Hanson, C. Zorrilla and J. F. Lew (1999). "Maternal levels of plasma human immunodeficiency virus type 1 RNA and the risk of perinatal transmission. Women and Infants Transmission Study Group." N Engl J Med **341**(6): 394-402.

Hamers, R. L., C. L. Wallis, C. Kityo, M. Siwale, K. Mandaliya, F. Conradie, M. E. Botes, M. Wellington, A. Osibogun, K. C. Sigaloff, I. Nankya, R. Schuurman, F. W. Wit, W. S. Stevens, M. van Vugt and T. F. de Wit (2011). "HIV-1 drug resistance in antiretroviral-naive individuals in sub-Saharan Africa after rollout of antiretroviral therapy: a multicentre observational study." Lancet Infect Dis **11**(10): 750-759.

Hill, A. (2013). "Optimizing HIV treatment." Curr Opin HIV AIDS **8**(1): 34-40.

Kobbe, R., S. Schalkwijk, G. Dunay, J. M. Eberhard, U. Schulze-Sturm, B. Hollwitz, O. Degen, M. Teulen, A. Colbers and D. Burger (2016). "Dolutegravir in breast milk and maternal and infant plasma during

breastfeeding." AIDS **30**(17): 2731-2733.
Manasa, J., D. Katzenstein, S. Cassol, M. L. Newell and T. de Oliveira (2012). "Primary drug resistance in South Africa: data from 10 years of surveys." AIDS Res Hum Retroviruses **28**(6): 558-565.
Mehta, U., C. Clerk, E. Allen, M. Yore, E. Sevene, J. Singlovic, M. Petzold, V. Mangiaterra, E. Elefant, F. M. Sullivan, L. B. Holmes and M. Gomes (2012). "Protocol for a drugs exposure pregnancy registry for implementation in resource-limited settings." BMC Pregnancy Childbirth **12**: 89.
Meintjes, G., J. Black and F. Conradie (2015). "Southern African HIV Clinicians Society adult antiretroviral therapy guidelines: Update on when to initiate antiretroviral therapy." S Afr Med J **16**(1): 4.
Meyers, K., H. Qian, Y. Wu, Y. Lao, Q. Chen, X. Dong, H. Li, Y. Yang, C. Jiang and Z. Zhou (2015). "Early Initiation of ARV During Pregnancy to Move towards Virtual Elimination of Mother-to-Child-Transmission of HIV-1 in Yunnan, China." PLoS One **10**(9): e0138104.
Musiime, V., F. Ssali, J. Kayiwa, W. Namala, H. Kizito, C. Kityo and P. Mugyenyi (2009). "Response to nonnucleoside reverse transcriptase inhibitor-based therapy in HIV-infected children with perinatal exposure to single-dose nevirapine." AIDS Res Hum Retroviruses **25**(10): 989-996.
Myer, L., T. K. Phillips, N. Y. Hsiao, A. Zerbe, G. Petro, L. G. Bekker, J. A. McIntyre and E. J. Abrams (2015). "Plasma viraemia in HIV-positive pregnant women entering antenatal care in South Africa." J Int AIDS Soc **18**: 20045.
Ndibazza, J., S. Lule, M. Nampijja, H. Mpairwe, G. Oduru, M. Kiggundu, M. Akello, L. Muhangi and A. M. Elliott (2011). "A description of congenital anomalies among infants in Entebbe, Uganda." Birth Defects Res A Clin Mol Teratol **91**(9): 857-861.
Olagunju, A., M. Siccardi, O. Okafor, O. Bolaji, S. Khoo and A. Owen (2014). Pharmacogenetics of Efavirenz Excretion into Human Breast Milk and Transfer to Breastfed Infants. 21st Conference on Retroviruses and Opportunistic Infections. Boston, USA.
Patel, N., S. Abdelsayed, M. Veve and C. D. Miller (2011). "Predictors of clinically significant drug-drug interactions among patients treated with nonnucleoside reverse transcriptase inhibitor-, protease inhibitor-, and raltegravir-based antiretroviral regimens." Ann Pharmacother **45**(3): 317-324.
Pilcher, C. D., C. Ospina-Norvell, A. Dasgupta, D. Jones, W. Hartogensis, S. Torres, F. Calderon, E. Demicco, E. Geng, M. Gandhi, D. V. Havlir and H. Hatano (2017). "The Effect of Same-Day Observed Initiation of Antiretroviral Therapy on HIV Viral Load and Treatment Outcomes in a US Public Health Setting." J Acquir Immune Defic Syndr **74**(1): 44-51.
Quinn, J. A., F. M. Munoz, B. Gonik, L. Frau, C. Cutland, T. Mallett-Moore, A. Kissou, F. Wittke, M. Das, T. Nunes, S. Pye, W. Watson, A. A. Ramos, J. F. Cordero, W. T. Huang, S. Kochhar, J. Buttery and G. Brighton Collaboration Preterm Birth Working (2016). "Preterm birth: Case definition & guidelines for data collection, analysis, and presentation of immunisation safety data." Vaccine **34**(49): 6047-6056.
Raffi, F., A. Rachlis, H. J. Stellbrink, W. D. Hardy, C. Torti, C. Orkin, M. Bloch, D. Podzamczer, V. Pokrovsky, F. Pulido, S. Almond, D. Margolis, C. Brennan and S. Min (2013). "Once-daily dolutegravir versus raltegravir in antiretroviral-naive adults with HIV-1 infection: 48 week results from the randomised, double-blind, non-inferiority SPRING-2 study." Lancet **381**(9868): 735-743.
Rosen, S., M. Maskew, M. P. Fox, C. Nyoni, C. Mongwenyana, G. Malete, I. Sanne, D. Bokaba, C. Sauls, J. Rohr and L. Long (2016). "Initiating Antiretroviral Therapy for HIV at a Patient's First Clinic Visit: The RapIT Randomized Controlled Trial." PLoS Med **13**(5): e1002015.
Sibiude, J., L. Madelbrot, S. Blanche, J. Le Chenadec, N. Boulbag-Bonnet, A. Faye, C. Dollfus, R. Tubiana, B. Khoshnood and J. Warszawski (2013). Birth defects and ART in the French Perinatal Cohort, a prospective exhaustive study among 13,124 life births from 1994 to 2010. Conference on Retorviruses and Opportunistic Inections. Atlanta, Georgia. **81**.
Suchard, M. S., E. Mayne, V. A. Green, S. Shalekoff, S. L. Donninger, W. S. Stevens, C. M. Gray and C. T. Tiemessen (2010). "FOXP3 expression is upregulated in CD4T cells in progressive HIV-1 infection and is a marker of disease severity." PLoS One **5**(7): e11762.
Tavares Da Silva, F., B. Gonik, M. McMillan, C. Keech, S. Dellicour, S. Bhange, M. Tila, D. M. Harper, C. Woods, A. T. Kawai, S. Kochhar, F. M. Munoz and G. Brighton Collaboration Stillbirth Working (2016). "Stillbirth: Case definition and guidelines for data collection, analysis, and presentation of maternal

immunization safety data." Vaccine **34**(49): 6057-6068.
Uganda, M. (2016). Consolidated guidelines for prevention and treatment of HIV in Uganda. Kampala, Uganda, The Republic of Uganda Ministry of Health.
van Lunzen, J., F. Maggiolo, J. R. Arribas, A. Rakhmanova, P. Yeni, B. Young, J. K. Rockstroh, S. Almond, I. Song, C. Brothers and S. Min (2012). "Once daily dolutegravir (S/GSK1349572) in combination therapy in antiretroviral-naive adults with HIV: planned interim 48 week results from SPRING-1, a dose-ranging, randomised, phase 2b trial." Lancet Infect Dis **12**(2): 111-118.
Vitoria, M., N. Ford, P. Clayden, A. L. Pozniak and A. M. Hill (2017). "When could new antiretrovirals be recommended for national treatment programmes in low-income and middle-income countries: results of a WHO Think Tank." Curr Opin HIV AIDS.
Walmsley, S., A. Antela, D. Clumeck, D. Duiculescu, A. Eberhard, F. Gutierrez, L. Hocqueloux, F. Maggiolo, U. Sandkowsky, C. Grainer, B. Wynne and K. Pappa (2012). Dolutegravir (DTG; S/GSK 1349572) + Abacavir/Lamivudine once daily statistically superior to Tenofovir/Emtricitabine/Efavirenz: 48-week results - SINGLE (ING114467). ICAAC. Glasgow.
Walmsley, S. L., A. Antela, N. Clumeck, D. Duiculescu, A. Eberhard, F. Gutierrez, L. Hocqueloux, F. Maggiolo, U. Sandkovsky, C. Granier, K. Pappa, B. Wynne, S. Min, G. Nichols and S. Investigators (2013). "Dolutegravir plus abacavir-lamivudine for the treatment of HIV-1 infection." N Engl J Med **369**(19): 1807-1818.
Westreich, D., D. Rubel, P. MacDonald, P. Majuba, M. Maskew, S. Nagar, I. Jaffray, P. MacPhail, S. Cole and I. Sanne (2010). Pregnancy, efavirenz, and birth outcomes in Johannesburg. 17th Conference on Retroviruses and Opportunistic Infections. San Diego.
WHO (2010). Antiretroviral drugs for treating pregnant women and preventing HIV infection in infants: towards universal access. Recommendations for a public health approach (2010 version). Geneva, World Health Organization.
WHO (2012). Technical update on treatment optimization. Use of efavirenz during pregnancy:a public health perspective. Geneva, World Health Organization.
WHO (2013). Consolidated guidelines on the use of antiretroviral drugs for treating and preventing HIV infection: recommendations for a public health approach. Geneva, World Health Organization.
Zhang, J., S. Hayes, B. M. Sadler, I. Minto, J. Brandt, S. Piscitelli, S. Min and I. H. Song (2015). "Population pharmacokinetics of dolutegravir in HIV-infected treatment-naive patients." Br J Clin Pharmacol **80**(3): 502-514.
Venter WDF, Moorhouse M, Sokhela S, et al. Dolutegravir plus Two Different Prodrugs of Tenofovir to Treat HIV. *N Engl J Med*. 2019;381(9):803-815. doi:10.1056/NEJMoa1902824
NAMSAL ANRS 12313 Study Group, Kouanfack C, Mpoudi-Etame M, et al. Dolutegravir-Based or Low-Dose Efavirenz-Based Regimen for the Treatment of HIV-1. *N Engl J Med*. 2019;381(9):816-826. doi:10.1056/NEJMoa1904340

20.PROTOCOL SIGNATURE PAGE

I agree to conduct the trial in accordance with ICH-GCP and the applicable regulatory requirements and with the approved protocol.
I agree to comply with the procedures for data recording/reporting
I agree to permit monitoring, auditing and inspection and to retain the trial related essential documentation for the period of time required according to ICH-GCP.

Name of Chief Investigator: Professor Saye Khoo

Signature:

Date:

21.APPENDICIES

APPENDIX 1 Safety Questionnaires
APPENDIX 2 Edinburgh Postnatal Depression Scale (EPDS)
APPENDIX 3 Hospital Anxiety and Depression Scale (HADS)
APPENDIX 4 Infant Gross Motor Screening Test (IGMST)
APPENDIX 5 DIADS Grading Scale
APPENDIX 6 Liverpool Causality Assessment Tool for Adverse Drug Reactions
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Appendix 1 Safety Questionnaires

a) Maternal Safety Questionnaire

Study ID Visit Date Visit code

Please ask the participant whether they have experienced any of the following, and circle 'Y' or 'N' as appropriate. If Y for medication please complete concomitant medication log

Nausea Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Vomiting Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Diarrhoea Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Fever Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Chills Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Sweating Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Skin rash Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Itch Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Chest pain Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Breathlessness Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Fast heart beat Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Headache Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Drowsiness Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Visual disturbance Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Hearing problems Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Dizziness Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Poor sleep Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Abnormal dreams Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Tiredness/fatigue Y N Start date __ / __ / __ End date __ / __ / __ Meds Y

N

Low mood Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Eyes turn yellow Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Dark urine Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Change in bowel habit Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Weakness (specify where) Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Other pain (specify where) Y N Start date __ / __ / __ End date __ / __ / __ Meds Y N

Other (state): _____

1) Have you taken your medication as prescribed? Y N
2) If not, why not? _____
3) Have you taken any new medication (including herbal/ traditional, over the counter and recreational/alcohol) since your last visit? Y N
4) If Y, please complete concomitant medication log
5) Have you any other new concerns about your health? _____

Form completed by

Participant initials

b) Infant Safety Questionnaire

Study Site Study ID Unique Number Visit Date

1) Have you any concerns about your infant's health? Y N

2) If Y, which of the following?

Poor feeding	Y	N
Irritability	Y	N
Lethargy	Y	N
Difficulty breathing	Y	N
Fast breathing	Y	N
Cough	Y	N
Skin rash	Y	N
Fever, chills	Y	N
Eyes turn yellow	Y	N
Dark urine	Y	N
Loose motions, diarrhoea	Y	N
Constipation	Y	N

Other (state): _____

Form completed by
Participant initials

Appendix 2 Edinburgh Postnatal Depression Scale

EPDS					
We would like to know how you have been feeling in the past week. Please choose the answer that comes closest to how you have felt in the past week, not just how you feel today. Please read all the options for each statement.					
		0	**1**	**2**	**3**
EPDS -1	I have been able to laugh and see the funny side of thing.	As much as I always could.	Not quite so much now.	Definitely not so much now.	Not at all.
EPDS -2	I have looked forward with enjoyment to things.	As much as I ever did.	A little less than I used to.	Much less than I used to.	Hardly at all.
EPDS -3	I have blamed myself unnecessarily when things went wrong.	Yes, most of the time.	Yes, some of the time.	Not very much.	No, never.
EPDS -4	I have been anxious or worried for no good reason.	No, not at all.	Hardly ever.	Yes, sometimes.	Yes, very much.
EPDS -5	I have felt scared or panicky for no very good reason.	Yes, quite a lot.	Yes, sometimes.	No, not much.	No, not at all.
EPDS -6	Things have been getting on top of me.	Yes, most of the times I haven't been managing at all.	Yes, sometimes I haven't been managing as well as usual.	No, most of the time I have managed quite well.	No, I have been managing as well as ever.
EPDS -7	I have been so unhappy that I have had difficulty sleeping.	Yes, most of the time.	Yes, sometimes.	Not very much.	No, not at all.
EPDS -8	I have felt sad or miserable.	Yes, most of the time.	Yes, sometimes.	Not very much.	No, not at all.
EPDS -9	I have been so unhappy that I have been crying.	Yes, most of the time.	Yes, sometimes.	Not very much.	No, not at all.
EPDS -10	The thought of harming myself has occurred to me.	Yes, quite a lot	Sometimes	Hardly ever	Never

Interpretation of Score

Edinburgh Postnatal Depression Scale (EPDS) [Cox, Holden & Sagovsky 1987]

The EPDS is a self-rated questionnaire that has been used in Europe and Australia for over 10 years to screen women for PPD. It asks women to rate how they have been feeling in the last 7 days and consists of 10 short statements of common depressive symptoms with 4 choices per statement. Each statement is rated on a scale of 0 – 3 with possible total scores ranging from 0 – 30.

To administer the test you give the woman a pen and the questionnaire and ask her to answer the questions in relation to the past 7 days. The questionnaire should only take a few minutes to complete.

Scoring the questionnaire only take a couple of minutes with practice.
Questions 3,5,6,7,8,9 and 10 are scored: statement 1 = 3 points, statement 2 = 2 points, statement 3 = 1 point and statement 4 = 0 points.

A cut-off score of 12.5 has been shown to detect major depression and a woman who meets this threshold can be further assessed. Asking a woman to complete such a questionnaire not only makes her stop and think about how she has been feeling but also indicates a willingness on the part of the person giving the questionnaire to listen to how she is feeling.

Appendix 3 Hospital Anxiety and Depression Scale (HADS)

D	A		D	A	
		I feel tense or 'wound up':			**I feel as if I am slowed down:**
	3	Most of the time	3		Nearly all the time
	2	A lot of the time	2		Very often
	1	From time to time, occasionally	1		Sometimes
	0	Not at all	0		Not at all
		I still enjoy the things I used to enjoy:			**I get a sort of frightened feeling like 'butterflies' in the stomach:**
0		Definitely as much		0	Not at all
1		Not quite so much		1	Occasionally
2		Only a little		2	Quite Often
3		Hardly at all		3	Very Often
		I get a sort of frightened feeling as if something awful is about to happen:			**I have lost interest in my appearance:**
	3	Very definitely and quite badly	3		Definitely
	2	Yes, but not too badly	2		I don't take as much care as I should
	1	A little, but it doesn't worry me	1		I may not take quite as much care
	0	Not at all	0		I take just as much care as ever
		I can laugh and see the funny side of things:			**I feel restless as I have to be on the move:**
0		As much as I always could		3	Very much indeed
1		Not quite so much now		2	Quite a lot
2		Definitely not so much now		1	Not very much
3		Not at all		0	Not at all
		Worrying thoughts go through my mind:			**I look forward with enjoyment to things:**
	3	A great deal of the time	0		As much as I ever did
	2	A lot of the time	1		Rather less than I used to
	1	From time to time, but not too often	2		Definitely less than I used to
	0	Only occasionally	3		Hardly at all

		I feel cheerful:			**I get sudden feelings of panic:**
3		Not at all		3	Very often indeed
2		Not often		2	Quite often
1		Sometimes		1	Not very often
0		Most of the time		0	Not at all
		I can sit at ease and feel relaxed:			**I can enjoy a good book or radio or TV program:**
	0	Definitely	0		Often
	1	Usually	1		Sometimes
	2	Not Often	2		Not often
	3	Not at all	3		Very seldom

Tick the box beside the reply that is closest to how you have been feeling in the past week.
Don't take too long over you replies: your immediate is best.

Please check you have answered all the questions

Scoring:

Total Score: Depression (D)_____ Anxiety (A)_____

0-7 = Normal

8-10 = Borderline abnormal (borderline case)

11-21 = Abnormal (case)

Appendix 4 Infant Gross Motor Screening Test (IGMST)

The Infant Gross Motor Screening Test 6-8 months

Date of Assessment____/____/_____

Child's name_____

Date of Birth / /_____

Age_____months

		Score
	Controls head at 90 degrees while lying on stomach Child lies on stomach, and should be able to lift head up to 90 degrees for about 5 seconds	☐
	Elevates chest whilst lying on stomach (with extended arms) Child supports weight on both hands, whilst keeping arms straight, and lifting head at 90 degrees	☐
	Plays with feet Child should bring one or both feet to hands (above the hips), and be able to maintain this position whilst playing with feet	☐
	Rolls from back to stomach Child can do this to both or one side. This should be voluntary, and initiated by lifting the head	☐
	Sits alone The child should be able to sit alone for about 30 seconds without using arms for support	☐

6-7 months	8 months
☐ 4-5 Satisfactory	☐ 5 Satisfactory
☐ 0-3 At risk	☐ 0-4 At risk

The Infant Gross Motor Screening Test 9-12 months

Child's name_____

Date of Birth / /_____

Date of Assessment____/____/_____

Age_____months

		Score
	**Turns body while seated** Child turns his or her trunk and reaches for object	☐
	**Makes stepping movements** Child makes at least two stepping movements that move him/herself forward whilst hands are held	☐
	**Moves from sitting to being on hands and knees** Child uses rotation to move from sitting to being on hands and knees	☐
	**Crawls/moves forward at least 1½ metres** Child uses crawling/moving on stomach and pulling with hands to move forwards at least 1 ½ metres	☐
	**Pulls up to standing position** Child uses an object such as a table in order to pull him/herself into a standing position, or pushes up on the floor without support	☐

9-10 months	11-12 months
☐ 4-5 Satisfactory	☐ 5 Satisfactory
☐ 0-3 At risk	☐ 0-4 At risk

The Infant Gross Motor Screening Test 13-16 months

Child's name_____

Date of Birth / /_____

Date of Assessment____/____/_____

Age_____months

		Score
	**Pulls up to standing position** Child uses an object such as a table in order to pull him/herself into a standing position, or pushes up on the floor without support	☐
	**Sits down from supported standing in a controlled manner** Child sits down from a standing position using good control e.g. by using a controlled squat	☐
	**Stands independently** Child is able to stand without support for at least 20 seconds	☐
	**Walks alone with coordination** Child should be able to walk a reasonable distance (such as 20 steps) with good control and coordination	☐
	**Squats without support** The child should be able to squat down with good control, and maintain this position for play. The child's bottom should not be resting on the floor	☐

13 months	14 months	15-16 months
☐ 3-5 Satisfactory	☐ 4-5 Satisfactory	☐ 5 Satisfactory
☐ 0-2 At risk	☐ 0-3 At risk	☐ 0-4 At risk

The Infant Gross Motor Screening Test 17-18 months

Child's name_____

Date of Birth / /_____

Date of Assessment____/____/_____

Age_____months

		Score
	**Sits down from supported standing in a controlled manner** Child sits down from a standing position using good control e.g. by using a controlled squat	☐
	**Stands independently** Child is able to stand without support for at least 1 minute	☐
	**Stands up with no assistance** Child moves into a standing position without using an object to pull on. The child may push up on the floor in order to stand up	☐
	**Walks alone with coordination** Child should be able to walk a reasonable distance (such as 40 steps) with good control and coordination	☐
	**Squats without support** The child should be able to squat down with good control, and maintain this position for play. The child's bottom should not be resting on the floor	☐
	**Runs with coordination** The child should be able to run without falling over	☐

17-18 months

☐ 6 Satisfactory

☐ 0-5 At risk

Appendix 5 DAIDS Grading Scale

Division of Aids (DAIDS) Table for Grading the Severity of Adult and Paediatric Adverse Events (Corrected V 2.1 July 2017)

The DAIDS grading table provides an AE severity grading scale ranging from grades 1 to 5 with descriptions for each AE based on the following general guidelines:

- Grade 1 indicates a mild event
- Grade 2 indicates a moderate event
- Grade 3 indicates a severe event
- Grade 4 indicates a potentially life-threatening event
- Grade 5 indicates death (*Note*: This grade is not specifically listed on each page of the grading table).

Other points to consider include:

- Use age and sex values as applicable.
- Unless noted, laboratory values are for term neonates. Preterm neonates should be assessed using local laboratory normal ranges.
- Where applicable, Standard International (SI) units are included in italics.

Selecting and Reporting a Primary AE Term

When selecting a primary AE term to report, sites should select the term that best describes what occurred to the participant. For example, a participant may present with itching, urticaria, flushing, angioedema of the face, and dyspnea. If the underlying diagnosis is determined to be an acute allergic reaction, sites should report "Acute Allergic Reaction" as the primary AE term.

Grading Adult and Pediatric AEs

When a single parameter is not appropriate for grading an AE in both adult and pediatric populations, separate parameters with specified age ranges are provided. If no distinction between adult and pediatric populations has been made, the listed parameter should be used for grading an AE in both populations.

Reporting Pregnancy Outcomes

In the *Pregnancy, Puerperium, and Perinatal* section, all parameters are pregnancy outcomes and should be reported using the mother's participant ID. If an infant is not enrolled in the same study as the mother, any identified birth defects should be reported using the mother's participant ID. However, if an infant is enrolled in the same study as the mother or in another study, any identified birth defects should be reported using the infant's participant ID. Sites should refer to the applicable network standards for reporting abnormal pregnancy outcomes on the CRFs.

Determining Severity Grade for Parameters between Grades

If the severity of an AE could fall in either one of two grades (i.e., the severity of an AE could be either grade 2 or grade 3), sites should select the higher of the two grades.

Laboratory Values

General. An asymptomatic, abnormal laboratory finding without an accompanying AE should not be reported to DAIDS in an expedited timeframe unless it meets protocol-specific reporting requirements. Sites should refer to the applicable network standards for reporting abnormal laboratory findings on the clinical case report forms.

Values below Grade 1. Any laboratory value that is between the ULN and grade 1 (for high values) or the LLN and grade 1 (for low values) should not be graded or reported as an AE. Sites should consult the

Manual for Expedited Reporting of Adverse Events to DAIDS, Version 2.0 and their protocol when making an assessment of the need to report an AE.

Overlap of Local Laboratory Normal Values with Grading Table Ranges.
When local laboratory normal values fall within grading table laboratory ranges, the severity grading is based on the ranges in the grading table unless there is a protocol-specific grading criterion for the laboratory value. For example, "Magnesium, Low" has a grade 1 range of 1.2 to < 1.4 mEq/L, while a particular laboratory's normal range for magnesium may be 1.3 to 2.8 mEq/L. If a study participant's magnesium laboratory value is 1.3 mEq/L, the laboratory value should be graded as grade 1.

Appendix Usage
Appendix A takes priority over the main grading table in all assessments of total bilirubin for term and preterm neonates.

Estimating Severity Grade for Parameters Not Identified in the Grading Table
The functional table below should be used to grade the severity of an AE that is not specifically identified in the grading table. In addition, all deaths related to an AE are to be classified as grade 5.

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Clinical adverse event **NOT** identified elsewhere in the grading table	Mild symptoms causing no or minimal interference with usual social & functional activities with intervention not indicated	Moderate symptoms causing greater than minimal interference with usual social & functional activities with intervention indicated	Severe symptoms causing inability to perform usual social & functional activities with intervention or hospitalization indicated	Potentially life-threatening symptoms causing inability to perform basic self-care functions with intervention indicated to prevent permanent impairment, persistent disability, or death

Major Clinical Conditions

Cardiovascular

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Arrhythmia (by ECG or physical examination) *Specify type, if applicable*	No symptoms AND No intervention indicated	No symptoms AND Non-urgent intervention indicated	Non-life-threatening symptoms AND Non-urgent intervention indicated	Life-threatening arrhythmia OR Urgent intervention indicated
Blood Pressure Abnormalities [1]				
Hypertension *(with the lowest reading taken after repeat testing during a visit)* *≥ 18 years of age*	140 to < 160 mmHg systolic OR 90 to < 100 mmHg diastolic	≥ 160 to < 180 mmHg systolic OR ≥ 100 to < 110 mmHg diastolic	≥ 180 mmHg systolic OR ≥ 110 mmHg diastolic	Life-threatening consequences in a participant not previously diagnosed with hypertension (e.g., malignant hypertension) OR Hospitalization indicated
< 18 years of age	> 120/80 mmHg	≥ 95th to < 99th percentile + 5 mmHg adjusted for age, height, and gender (systolic and/or diastolic)	≥ 99th percentile + 5 mmHg adjusted for age, height, and gender (systolic and/or diastolic)	Life-threatening consequences in a participant not previously diagnosed with hypertension (e.g., malignant hypertension) OR Hospitalization indicated
Hypotension	No symptoms	Symptoms corrected with oral fluid replacement	Symptoms AND IV fluids indicated	Shock requiring use of vasopressors or mechanical assistance to maintain blood pressure
Cardiac Ischemia or Infarction *Report only one*	NA	NA	New symptoms with ischemia (stable angina) OR New testing consistent with ischemia	Unstable angina OR Acute myocardial infarction
Heart Failure	No symptoms AND Laboratory or cardiac imaging abnormalities	Symptoms with mild to moderate activity or exertion	Symptoms at rest or with minimal activity or exertion (e.g., hypoxemia) OR Intervention indicated (e.g., oxygen)	Life-threatening consequences OR Urgent intervention indicated (e.g., vasoactive medications, ventricular assist device, heart transplant)

[1] Blood pressure norms for children < 18 years of age can be found in: Expert Panel on Integrated Guidelines for Cardiovascular Health and Risk Reduction in Children and Adolescents. *Pediatrics* 2011;128;S213; originally published online November 14, 2011; DOI: 10.1542/peds.2009-2107C.

Cardiovascular

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Hemorrhage (with significant acute blood loss)	NA	Symptoms AND No transfusion indicated	Symptoms AND Transfusion of ≤ 2 units packed RBCs indicated	Life-threatening hypotension OR Transfusion of > 2 units packed RBCs (for children, packed RBCs > 10 cc/kg) indicated
Prolonged PR Interval or AV Block *Report only one* *> 16 years of age*	PR interval 0.21 to < 0.25 seconds	PR interval ≥ 0.25 seconds OR Type I 2nd degree AV block	Type II 2nd degree AV block OR Ventricular pause ≥ 3.0 seconds	Complete AV block
≤ 16 years of age	1st degree AV block (PR interval > normal for age and rate)	Type I 2nd degree AV block	Type II 2nd degree AV block OR Ventricular pause ≥ 3.0 seconds	Complete AV block
Prolonged QTc Interval[2]	0.45 to 0.47 seconds	> 0.47 to 0.50 seconds	> 0.50 seconds OR ≥ 0.06 seconds above baseline	Life-threatening consequences (e.g., Torsade de pointes, other associated serious ventricular dysrhythmia)
Thrombosis or Embolism *Report only one*	NA	Symptoms AND No intervention indicated	Symptoms AND Intervention indicated	Life-threatening embolic event (e.g., pulmonary embolism, thrombus)

[2] As per Bazett's formula.

Dermatologic

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Alopecia (scalp only)	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing greater than minimal interference with usual social & functional activities	NA	NA
Bruising	Localized to one area	Localized to more than one area	Generalized	NA
Cellulitis	NA	Non-parenteral treatment indicated (e.g., oral antibiotics, antifungals, antivirals)	IV treatment indicated (e.g., IV antibiotics, antifungals, antivirals)	Life-threatening consequences (e.g., sepsis, tissue necrosis)
Hyperpigmentation	Slight or localized causing no or minimal interference with usual social & functional activities	Marked or generalized causing greater than minimal interference with usual social & functional activities	NA	NA
Hypopigmentation	Slight or localized causing no or minimal interference with usual social & functional activities	Marked or generalized causing greater than minimal interference with usual social & functional activities	NA	NA
Petechiae	Localized to one area	Localized to more than one area	Generalized	NA
Pruritus [3] (without skin lesions)	Itching causing no or minimal interference with usual social & functional activities	Itching causing greater than minimal interference with usual social & functional activities	Itching causing inability to perform usual social & functional activities	NA
Rash *Specify type, if applicable*	Localized rash	Diffuse rash OR Target lesions	Diffuse rash AND Vesicles or limited number of bullae or superficial ulcerations of mucous membrane limited to one site	Extensive or generalized bullous lesions OR Ulceration of mucous membrane involving two or more distinct mucosal sites OR Stevens-Johnson syndrome OR Toxic epidermal necrolysis

[3] For pruritus associated with injections or infusions, see the *Site Reactions to Injections and Infusions* section (page 23).

Endocrine and Metabolic

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Diabetes Mellitus	Controlled without medication	Controlled with medication OR Modification of current medication regimen	Uncontrolled despite treatment modification OR Hospitalization for immediate glucose control indicated	Life-threatening consequences (e.g., ketoacidosis, hyperosmolar non-ketotic coma, end organ failure)
Gynecomastia	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing pain with greater than minimal interference with usual social & functional activities	Disfiguring changes AND Symptoms requiring intervention or causing inability to perform usual social & functional activities	NA
Hyperthyroidism	No symptoms AND Abnormal laboratory value	Symptoms causing greater than minimal interference with usual social & functional activities OR Thyroid suppression therapy indicated	Symptoms causing inability to perform usual social & functional activities OR Uncontrolled despite treatment modification	Life-threatening consequences (e.g., thyroid storm)
Hypothyroidism	No symptoms AND Abnormal laboratory value	Symptoms causing greater than minimal interference with usual social & functional activities OR Thyroid replacement therapy indicated	Symptoms causing inability to perform usual social & functional activities OR Uncontrolled despite treatment modification	Life-threatening consequences (e.g., myxedema coma)
Lipoatrophy[4]	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing greater than minimal interference with usual social & functional activities	Disfiguring changes	NA

[4] Definition: A disorder characterized by fat loss in the face, extremities, and buttocks.

Endocrine and Metabolic

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Lipohypertrophy[5]	Detectable by study participant, caregiver, or physician AND Causing no or minimal interference with usual social & functional activities	Obvious on visual inspection AND Causing greater than minimal interference with usual social & functional activities	Disfiguring changes	NA

[5] Definition: A disorder characterized by abnormal fat accumulation on the back of the neck, breasts, and abdomen.

Gastrointestinal

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Anorexia	Loss of appetite without decreased oral intake	Loss of appetite associated with decreased oral intake without significant weight loss	Loss of appetite associated with significant weight loss	Life-threatening consequences OR Aggressive intervention indicated (e.g., tube feeding, total parenteral nutrition)
Ascites	No symptoms	Symptoms AND Intervention indicated (e.g., diuretics, therapeutic paracentesis)	Symptoms recur or persist despite intervention	Life-threatening consequences
Bloating or Distension *Report only one*	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	NA
Cholecystitis	NA	Symptoms AND Medical intervention indicated	Radiologic, endoscopic, or operative intervention indicated	Life-threatening consequences (e.g., sepsis, perforation)
Constipation	NA	Persistent constipation requiring regular use of dietary modifications, laxatives, or enemas	Obstipation with manual evacuation indicated	Life-threatening consequences (e.g., obstruction)
Diarrhea *≥ 1 year of age*	Transient or intermittent episodes of unformed stools OR Increase of ≤ 3 stools over baseline per 24-hour period	Persistent episodes of unformed to watery stools OR Increase of 4 to 6 stools over baseline per 24-hour period	Increase of ≥ 7 stools per 24-hour period OR IV fluid replacement indicated	Life-threatening consequences (e.g., hypotensive shock)
< 1 year of age	Liquid stools (more unformed than usual) but usual number of stools	Liquid stools with increased number of stools OR Mild dehydration	Liquid stools with moderate dehydration	Life-threatening consequences (e.g., liquid stools resulting in severe dehydration, hypotensive shock)
Dysphagia or Odynophagia *Report only one and specify location*	Symptoms but able to eat usual diet	Symptoms causing altered dietary intake with no intervention indicated	Symptoms causing severely altered dietary intake with intervention indicated	Life-threatening reduction in oral intake
Gastrointestinal Bleeding	Not requiring intervention other than iron supplement	Endoscopic intervention indicated	Transfusion indicated	Life-threatening consequences (e.g., hypotensive shock)

Gastrointestinal

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Mucositis or Stomatitis *Report only one and specify location*	Mucosal erythema	Patchy pseudomembranes or ulcerations	Confluent pseudomembranes or ulcerations OR Mucosal bleeding with minor trauma	Life-threatening consequences (e.g., aspiration, choking) OR Tissue necrosis OR Diffuse spontaneous mucosal bleeding
Nausea	Transient (< 24 hours) or intermittent AND No or minimal interference with oral intake	Persistent nausea resulting in decreased oral intake for 24 to 48 hours	Persistent nausea resulting in minimal oral intake for > 48 hours OR Rehydration indicated (e.g., IV fluids)	Life-threatening consequences (e.g., hypotensive shock)
Pancreatitis	NA	Symptoms with hospitalization not indicated	Symptoms with hospitalization indicated	Life-threatening consequences (e.g., circulatory failure, hemorrhage, sepsis)
Perforation (colon or rectum)	NA	NA	Intervention indicated	Life-threatening consequences
Proctitis	Rectal discomfort with no intervention indicated	Symptoms causing greater than minimal interference with usual social & functional activities OR Medical intervention indicated	Symptoms causing inability to perform usual social & functional activities OR Operative intervention indicated	Life-threatening consequences (e.g., perforation)
Rectal Discharge	Visible discharge	Discharge requiring the use of pads	NA	NA
Vomiting	Transient or intermittent AND No or minimal interference with oral intake	Frequent episodes with no or mild dehydration	Persistent vomiting resulting in orthostatic hypotension OR Aggressive rehydration indicated (e.g., IV fluids)	Life-threatening consequences (e.g., hypotensive shock)

Musculoskeletal

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Arthralgia	Joint pain causing no or minimal interference with usual social & functional activities	Joint pain causing greater than minimal interference with usual social & functional activities	Joint pain causing inability to perform usual social & functional activities	Disabling joint pain causing inability to perform basic self-care functions
Arthritis	Stiffness or joint swelling causing no or minimal interference with usual social & functional activities	Stiffness or joint swelling causing greater than minimal interference with usual social & functional activities	Stiffness or joint swelling causing inability to perform usual social & functional activities	Disabling joint stiffness or swelling causing inability to perform basic self-care functions
Myalgia (generalized)	Muscle pain causing no or minimal interference with usual social & functional activities	Muscle pain causing greater than minimal interference with usual social & functional activities	Muscle pain causing inability to perform usual social & functional activities	Disabling muscle pain causing inability to perform basic self-care functions
Osteonecrosis	NA	No symptoms but with radiographic findings AND No operative intervention indicated	Bone pain with radiographic findings OR Operative intervention indicated	Disabling bone pain with radiographic findings causing inability to perform basic self-care functions
Osteopenia[6] *≥ 30 years of age*	BMD t-score -2.5 to -1	NA	NA	NA
< 30 years of age	BMD z-score -2 to -1	NA	NA	NA
Osteoporosis[6] *≥ 30 years of age*	NA	BMD t-score < -2.5	Pathologic fracture (e.g., compression fracture causing loss of vertebral height)	Pathologic fracture causing life-threatening consequences
< 30 years of age	NA	BMD z-score < -2	Pathologic fracture (e.g., compression fracture causing loss of vertebral height)	Pathologic fracture causing life-threatening consequences

[6] BMD t and z scores can be found in: Kanis JA on behalf of the World Health Organization Scientific Group (2007). Assessment of osteoporosis at the primary health-care level. Technical Report. World Health Organization Collaborating Centre for Metabolic Bone Diseases, University of Sheffield, UK. 2007: Printed by the University of Sheffield.

Neurologic

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Acute CNS Ischemia	NA	NA	Transient ischemic attack	Cerebral vascular accident (e.g., stroke with neurological deficit)
Altered Mental Status (for Dementia, see *Cognitive, Behavioral, or Attentional Disturbance* below)	Changes causing no or minimal interference with usual social & functional activities	Mild lethargy or somnolence causing greater than minimal interference with usual social & functional activities	Confusion, memory impairment, lethargy, or somnolence causing inability to perform usual social & functional activities	Delirium OR Obtundation OR Coma
Ataxia	Symptoms causing no or minimal interference with usual social & functional activities OR No symptoms with ataxia detected on examination	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	Disabling symptoms causing inability to perform basic self-care functions
Cognitive, Behavioral, or Attentional Disturbance (includes dementia and attention deficit disorder) *Specify type, if applicable*	Disability causing no or minimal interference with usual social & functional activities OR Specialized resources not indicated	Disability causing greater than minimal interference with usual social & functional activities OR Specialized resources on part-time basis indicated	Disability causing inability to perform usual social & functional activities OR Specialized resources on a full-time basis indicated	Disability causing inability to perform basic self-care functions OR Institutionalization indicated
Developmental Delay *< 18 years of age* *Specify type, if applicable*	Mild developmental delay, either motor or cognitive, as determined by comparison with a developmental screening tool appropriate for the setting	Moderate developmental delay, either motor or cognitive, as determined by comparison with a developmental screening tool appropriate for the setting	Severe developmental delay, either motor or cognitive, as determined by comparison with a developmental screening tool appropriate for the setting	Developmental regression, either motor or cognitive, as determined by comparison with a developmental screening tool appropriate for the setting
Headache	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	Symptoms causing inability to perform basic self-care functions OR Hospitalization indicated OR Headache with significant impairment of alertness or other neurologic function

Neurologic

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Neuromuscular Weakness (includes myopathy and neuropathy) *Specify type, if applicable*	Minimal muscle weakness causing no or minimal interference with usual social & functional activities OR No symptoms with decreased strength on examination	Muscle weakness causing greater than minimal interference with usual social & functional activities	Muscle weakness causing inability to perform usual social & functional activities	Disabling muscle weakness causing inability to perform basic self-care functions OR Respiratory muscle weakness impairing ventilation
Neurosensory Alteration (includes paresthesia and painful neuropathy) *Specify type, if applicable*	Minimal paresthesia causing no or minimal interference with usual social & functional activities OR No symptoms with sensory alteration on examination	Sensory alteration or paresthesia causing greater than minimal interference with usual social & functional activities	Sensory alteration or paresthesia causing inability to perform usual social & functional activities	Disabling sensory alteration or paresthesia causing inability to perform basic self-care functions
Seizures ***New Onset Seizure*** *≥ 18 years of age*	NA	NA	1 to 3 seizures	Prolonged and repetitive seizures (e.g., status epilepticus) OR Difficult to control (e.g., refractory epilepsy)
< 18 years of age (includes new or pre-existing febrile seizures)	Seizure lasting < 5 minutes with < 24 hours postictal state	Seizure lasting 5 to < 20 minutes with < 24 hours postictal state	Seizure lasting ≥ 20 minutes OR > 24 hours postictal state	Prolonged and repetitive seizures (e.g., status epilepticus) OR Difficult to control (e.g., refractory epilepsy)
Pre-existing Seizure	NA	Increased frequency from previous level of control without change in seizure character	Change in seizure character either in duration or quality (e.g., severity or focality)	Prolonged and repetitive seizures (e.g., status epilepticus) OR Difficult to control (e.g., refractory epilepsy)
Syncope	Near syncope without loss of consciousness (e.g., pre-syncope)	Loss of consciousness with no intervention indicated	Loss of consciousness AND Hospitalization or intervention required	NA

Pregnancy, Puerperium, and Perinatal

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Stillbirth (report using mother's participant ID) *Report only one*	NA	NA	Fetal death occurring at ≥ 20 weeks gestation	NA
Preterm Birth (report using mother's participant ID)	Live birth at 34 to < 37 weeks gestational age	Live birth at 28 to < 34 weeks gestational age	Live birth at 24 to < 28 weeks gestational age	Live birth at < 24 weeks gestational age
Spontaneous Abortion or Miscarriage[7] (report using mother's participant ID) *Report only one*	Chemical pregnancy	Uncomplicated spontaneous abortion or miscarriage	Complicated spontaneous abortion or miscarriage	NA

[7] Definition: A pregnancy loss occurring at < 20 weeks gestational age.

Psychiatric

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Insomnia	Mild difficulty falling asleep, staying asleep, or waking up early causing no or minimal interference with usual social & functional activities	Moderate difficulty falling asleep, staying asleep, or waking up early causing more than minimal interference with usual social & functional activities	Severe difficulty falling asleep, staying asleep, or waking up early causing inability to perform usual social & functional activities requiring intervention or hospitalization	NA
Psychiatric Disorders (includes anxiety, depression, mania, and psychosis) *Specify disorder*	Symptoms with intervention not indicated OR Behavior causing no or minimal interference with usual social & functional activities	Symptoms with intervention indicated OR Behavior causing greater than minimal interference with usual social & functional activities	Symptoms with hospitalization indicated OR Behavior causing inability to perform usual social & functional activities	Threatens harm to self or others OR Acute psychosis OR Behavior causing inability to perform basic self-care functions
Suicidal Ideation or Attempt *Report only one*	Preoccupied with thoughts of death AND No wish to kill oneself	Preoccupied with thoughts of death AND Wish to kill oneself with no specific plan or intent	Thoughts of killing oneself with partial or complete plans but no attempt to do so OR Hospitalization indicated	Suicide attempted

Respiratory

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Acute Bronchospasm	Forced expiratory volume in 1 second or peak flow reduced to ≥ 70 to < 80% OR Mild symptoms with intervention not indicated	Forced expiratory volume in 1 second or peak flow 50 to < 70% OR Symptoms with intervention indicated OR Symptoms causing greater than minimal interference with usual social & functional activities	Forced expiratory volume in 1 second or peak flow 25 to < 50% OR Symptoms causing inability to perform usual social & functional activities	Forced expiratory volume in 1 second or peak flow < 25% OR Life-threatening respiratory or hemodynamic compromise OR Intubation
Dyspnea or Respiratory Distress *Report only one*	Dyspnea on exertion with no or minimal interference with usual social & functional activities OR Wheezing OR Minimal increase in respiratory rate for age	Dyspnea on exertion causing greater than minimal interference with usual social & functional activities OR Nasal flaring OR Intercostal retractions OR Pulse oximetry 90 to < 95%	Dyspnea at rest causing inability to perform usual social & functional activities OR Pulse oximetry < 90%	Respiratory failure with ventilator support indicated (e.g., CPAP, BPAP, intubation)

Sensory

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Hearing Loss *≥ 12 years of age*	NA	Hearing aid or intervention not indicated	Hearing aid or intervention indicated	Profound bilateral hearing loss (> 80 dB at 2 kHz and above) OR Non-serviceable hearing (i.e., >50 dB audiogram and <50% speech discrimination)
< 12 years of age (based on a 1, 2, 3, 4, 6 and 8 kHz audiogram)	> 20 dB hearing loss at ≤ 4 kHz	> 20 dB hearing loss at > 4 kHz	> 20 dB hearing loss at ≥ 3 kHz in one ear with additional speech language related services indicated (where available) OR Hearing loss sufficient to indicate therapeutic intervention, including hearing aids	Audiologic indication for cochlear implant and additional speech-language related services indicated (where available)
Tinnitus	Symptoms causing no or minimal interference with usual social & functional activities with intervention not indicated	Symptoms causing greater than minimal interference with usual social & functional activities with intervention indicated	Symptoms causing inability to perform usual social & functional activities	NA
Uveitis	No symptoms AND Detectable on examination	Anterior uveitis with symptoms OR Medical intervention indicated	Posterior or pan-uveitis OR Operative intervention indicated	Disabling visual loss in affected eye(s)
Vertigo	Vertigo causing no or minimal interference with usual social & functional activities	Vertigo causing greater than minimal interference with usual social & functional activities	Vertigo causing inability to perform usual social & functional activities	Disabling vertigo causing inability to perform basic self-care functions
Visual Changes (assessed from baseline)	Visual changes causing no or minimal interference with usual social & functional activities	Visual changes causing greater than minimal interference with usual social & functional activities	Visual changes causing inability to perform usual social & functional activities	Disabling visual loss in affected eye(s)

Systemic

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Acute Allergic Reaction	Localized urticaria (wheals) with no medical intervention indicated	Localized urticaria with intervention indicated OR Mild angioedema with no intervention indicated	Generalized urticaria OR Angioedema with intervention indicated OR Symptoms of mild bronchospasm	Acute anaphylaxis OR Life-threatening bronchospasm OR Laryngeal edema
Chills	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	NA
Cytokine Release Syndrome[8]	Mild signs and symptoms AND Therapy (i.e., antibody infusion) interruption not indicated	Therapy (i.e., antibody infusion) interruption indicated AND Responds promptly to symptomatic treatment OR Prophylactic medications indicated for ≤ 24 hours	Prolonged severe signs and symptoms OR Recurrence of symptoms following initial improvement	Life-threatening consequences (e.g., requiring pressor or ventilator support)
Fatigue or Malaise *Report only one*	Symptoms causing no or minimal interference with usual social & functional activities	Symptoms causing greater than minimal interference with usual social & functional activities	Symptoms causing inability to perform usual social & functional activities	Incapacitating symptoms of fatigue or malaise causing inability to perform basic self-care functions
Fever (non-axillary temperatures only)	38.0 to < 38.6°C or 100.4 to < 101.5°F	≥ 38.6 to < 39.3°C or ≥ 101.5 to < 102.7°F	≥ 39.3 to < 40.0°C or ≥ 102.7 to < 104.0°F	≥ 40.0°C or ≥ 104.0°F
Pain[9] (not associated with study agent injections and not specified elsewhere) *Specify location*	Pain causing no or minimal interference with usual social & functional activities	Pain causing greater than minimal interference with usual social & functional activities	Pain causing inability to perform usual social & functional activities	Disabling pain causing inability to perform basic self-care functions OR Hospitalization indicated

[8] Definition: A disorder characterized by nausea, headache, tachycardia, hypotension, rash, and/or shortness of breath.
[9] For pain associated with injections or infusions, see the *Site Reactions to Injections and Infusions* section.

Systemic

Serum Sickness[10]	Mild signs and symptoms	Moderate signs and symptoms AND Intervention indicated (e.g., antihistamines)	Severe signs and symptoms AND Higher level intervention indicated (e.g., steroids or IV fluids)	Life-threatening consequences (e.g., requiring pressor or ventilator support)
Underweight[11] *> 5 to 19 years of age*	WHO BMI z-score < -1 to -2	WHO BMI z-score < -2 to -3	WHO BMI z-score < -3	WHO BMI z-score < -3 with life-threatening consequences
2 to 5 years of age	WHO Weight-for-height z-score < -1 to -2	WHO Weight-for-height z-score < -2 to -3	WHO Weight-for-height z-score < -3	WHO Weight-for-height z-score < -3 with life-threatening consequences
< 2 years of age	WHO Weight-for-length z-score < -1 to -2	WHO Weight-for-length z-score < -2 to -3	WHO Weight-for-length z-score < -3	WHO Weight-for-length z-score < -3 with life-threatening consequences
Unintentional Weight Loss (excludes postpartum weight loss)	NA	5 to < 9% loss in body weight from baseline	≥ 9 to < 20% loss in body weight from baseline	≥ 20% loss in body weight from baseline OR Aggressive intervention indicated (e.g., tube feeding, total parenteral nutrition)

[10] Definition: A disorder characterized by fever, arthralgia, myalgia, skin eruptions, lymphadenopathy, marked discomfort, and/or dyspnea.

[11] WHO reference tables may be accessed by clicking the desired age range or by accessing the following URLs: http://www.who.int/growthref/who2007_bmi_for_age/en/ for participants > 5 to 19 years of age and http://www.who.int/childgrowth/standards/chart_catalogue/en/ for those ≤ 5 years of age.

Urinary

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Urinary Tract Obstruction	NA	Signs or symptoms of urinary tract obstruction without hydronephrosis or renal dysfunction	Signs or symptoms of urinary tract obstruction with hydronephrosis or renal dysfunction	Obstruction causing life-threatening consequences

Site Reactions to Injections and Infusions

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Injection Site Pain or Tenderness *Report only one*	Pain or tenderness causing no or minimal limitation of use of limb	Pain or tenderness causing greater than minimal limitation of use of limb	Pain or tenderness causing inability to perform usual social & functional activities	Pain or tenderness causing inability to perform basic self-care function OR Hospitalization indicated
Injection Site Erythema or Redness [12] *Report only one* *> 15 years of age*	2.5 to < 5 cm in diameter OR 6.25 to < 25 cm^2 surface area AND Symptoms causing no or minimal interference with usual social & functional activities	≥ 5 to < 10 cm in diameter OR ≥ 25 to < 100 cm^2 surface area OR Symptoms causing greater than minimal interference with usual social & functional activities	≥ 10 cm in diameter OR ≥ 100 cm^2 surface area OR Ulceration OR Secondary infection OR Phlebitis OR Sterile abscess OR Drainage OR Symptoms causing inability to perform usual social & functional activities	Potentially life-threatening consequences (e.g., abscess, exfoliative dermatitis, necrosis involving dermis or deeper tissue)
≤ 15 years of age	≤ 2.5 cm in diameter	> 2.5 cm in diameter with < 50% surface area of the extremity segment involved (e.g., upper arm or thigh)	≥ 50% surface area of the extremity segment involved (e.g., upper arm or thigh) OR Ulceration OR Secondary infection OR Phlebitis OR Sterile abscess OR Drainage	Potentially life-threatening consequences (e.g., abscess, exfoliative dermatitis, necrosis involving dermis or deeper tissue)
Injection Site Induration or Swelling *Report only one* *> 15 years of age*	Same as for **Injection Site Erythema or Redness,** *> 15 years of age*	Same as for **Injection Site Erythema or Redness,** *> 15 years of age*	Same as for **Injection Site Erythema or Redness,** *> 15 years of age*	Same as for **Injection Site Erythema or Redness,** *> 15 years of age*
≤ 15 years of age	Same as for **Injection Site Erythema or Redness,** *≤ 15 years of age*	Same as for **Injection Site Erythema or Redness,** *≤ 15 years of age*	Same as for **Injection Site Erythema or Redness,** *≤ 15 years of age*	Same as for **Injection Site Erythema or Redness,** *≤ 15 years of age*
Injection Site Pruritus	Itching localized to the injection site that is relieved spontaneously or in < 48 hours of treatment	Itching beyond the injection site that is not generalized OR Itching localized to the injection site requiring ≥ 48 hours treatment	Generalized itching causing inability to perform usual social & functional activities	NA

[12] Injection Site Erythema or Redness should be evaluated and graded using the greatest single diameter or measured surface area.

*Laboratory Values**

Chemistries

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Acidosis	NA	pH ≥ 7.3 to < LLN	pH < 7.3 without life-threatening consequences	pH < 7.3 with life-threatening consequences
Albumin, Low (g/dL; *g/L*)	3.0 to < LLN *30 to < LLN*	≥ 2.0 to < 3.0 *≥ 20 to < 30*	< 2.0 *< 20*	NA
Alkaline Phosphatase, High	1.25 to < 2.5 x ULN	2.5 to < 5.0 x ULN	5.0 to < 10.0 x ULN	≥ 10.0 x ULN
Alkalosis	NA	pH > ULN to ≤ 7.5	pH > 7.5 without life-threatening consequences	pH > 7.5 with life-threatening consequences
ALT or SGPT, High *Report only one*	1.25 to < 2.5 x ULN	2.5 to < 5.0 x ULN	5.0 to < 10.0 x ULN	≥ 10.0 x ULN
Amylase (Pancreatic) or Amylase (Total), High *Report only one*	1.1 to < 1.5 x ULN	1.5 to < 3.0 x ULN	3.0 to < 5.0 x ULN	≥ 5.0 x ULN
AST or SGOT, High *Report only one*	1.25 to < 2.5 x ULN	2.5 to < 5.0 x ULN	5.0 to < 10.0 x ULN	≥ 10.0 x ULN
Bicarbonate, Low (mEq/L; *mmol/L*)	16.0 to < LLN *16.0 to < LLN*	11.0 to < 16.0 *11.0 to < 16.0*	8.0 to < 11.0 *8.0 to < 11.0*	< 8.0 *< 8.0*
Bilirubin ***Direct Bilirubin[13], High*** *> 28 days of age*	NA	NA	> ULN with other signs and symptoms of hepatotoxicity.	> ULN with life-threatening consequences (e.g., signs and symptoms of liver failure)
≤ 28 days of age	ULN to ≤ 1 mg/dL	> 1 to ≤ 1.5 mg/dL	> 1.5 to ≤ 2 mg/dL	> 2 mg/dL
Total Bilirubin, High *> 28 days of age*	1.1 to < 1.6 x ULN	1.6 to < 2.6 x ULN	2.6 to < 5.0 x ULN	≥ 5.0 x ULN
≤ 28 days of age	See Appendix A. Total Bilirubin for Term and Preterm Neonates	See Appendix A. Total Bilirubin for Term and Preterm Neonates	See Appendix A. Total Bilirubin for Term and Preterm Neonates	See Appendix A. Total Bilirubin for Term and Preterm Neonates

*Reminder: An asymptomatic abnormal laboratory finding without an accompanying AE should not be reported to DAIDS in an expedited time frame unless it meets protocol-specific reporting requirements.

[13] Direct bilirubin > 1.5 mg/dL in a participant < 28 days of age should be graded as grade 2, if < 10% of the total bilirubin.

Chemistries

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Calcium, High (mg/dL; *mmol/L*)				
≥ *7 days of age*	10.6 to < 11.5 *2.65 to < 2.88*	11.5 to < 12.5 *2.88 to < 3.13*	12.5 to < 13.5 *3.13 to < 3.38*	≥ 13.5 *≥ 3.38*
< 7 days of age	11.5 to < 12.4 *2.88 to < 3.10*	12.4 to < 12.9 *3.10 to < 3.23*	12.9 to < 13.5 *3.23 to < 3.38*	≥ 13.5 *≥ 3.38*
Calcium (Ionized), High (mg/dL; *mmol/L*)	> ULN to < 6.0 *> ULN to < 1.5*	6.0 to < 6.4 *1.5 to < 1.6*	6.4 to < 7.2 *1.6 to < 1.8*	≥ 7.2 *≥ 1.8*
Calcium, Low (mg/dL; *mmol/L*)				
≥ *7 days of age*	7.8 to < 8.4 *1.95 to < 2.10*	7.0 to < 7.8 *1.75 to < 1.95*	6.1 to < 7.0 *1.53 to < 1.75*	< 6.1 *< 1.53*
< 7 days of age	6.5 to < 7.5 *1.63 to < 1.88*	6.0 to < 6.5 *1.50 to < 1.63*	5.50 to < 6.0 *1.38 to < 1.50*	< 5.50 *< 1.38*
Calcium (Ionized), Low (mg/dL; *mmol/L*)	< LLN to 4.0 *< LLN to 1.0*	3.6 to < 4.0 *0.9 to < 1.0*	3.2 to < 3.6 *0.8 to < 0.9*	< 3.2 *< 0.8*
Cardiac Troponin I, High	NA	NA	NA	Levels consistent with myocardial infarction or unstable angina as defined by the local laboratory
Creatine Kinase, High	3 to < 6 x ULN	6 to < 10x ULN	10 to < 20 x ULN	≥ 20 x ULN
Creatinine, High **Report only one*	1.1 to 1.3 x ULN	> 1.3 to 1.8 x ULN OR Increase to 1.3 to < 1.5 x participant's baseline	> 1.8 to < 3.5 x ULN OR Increase to 1.5 to < 2.0 x participant's baseline	≥ 3.5 x ULN OR Increase of ≥ 2.0 x participant's baseline
Creatinine Clearance [14] or eGFR, Low **Report only one*	NA	< 90 to 60 ml/min or ml/min/1.73 m^2 OR 10 to < 30% decrease from participant's baseline	< 60 to 30 ml/min or ml/min/1.73 m^2 OR 30 to < 50% decrease from participant's baseline	< 30 ml/min or ml/min/1.73 m^2 OR ≥ 50% decrease from participant's baseline or dialysis needed
Glucose (mg/dL; *mmol/L*)				
Fasting, High	110 to 125 *6.11 to < 6.95*	> 125 to 250 *6.95 to < 13.89*	> 250 to 500 *13.89 to < 27.75*	≥ 500 *≥ 27.75*
Nonfasting, High	116 to 160 *6.44 to < 8.89*	> 160 to 250 *8.89 to < 13.89*	> 250 to 500 *13.89 to < 27.75*	≥ 500 *≥ 27.75*

[14] Use the applicable formula (i.e., Cockcroft-Gault in mL/min or Schwartz, MDRD, CKD-Epi in mL/min/1.73m2). Sites should choose the method defined in their study and when not specified, use the method most relevant to the study population.
*Reminder: Choose the method that selects for the higher grade.

Chemistries

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Glucose, Low (mg/dL; *mmol/L*)				
≥ 1 month of age	55 to 64 *3.05 to <3.55*	40 to < 55 *2.22 to < 3.05*	30 to < 40 *1.67 to < 2.22*	< 30 *< 1.67*
< 1 month of age	50 to 54 *2.78 to < 3.00*	40 to < 50 *2.22 to < 2.78*	30 to < 40 *1.67 to < 2.22*	< 30 *< 1.67*
Lactate, High	ULN to < 2.0 x ULN without acidosis	≥ 2.0 x ULN without acidosis	Increased lactate with pH < 7.3 without life-threatening consequences	Increased lactate with pH < 7.3 with life-threatening consequences
Lipase, High	1.1 to < 1.5 x ULN	1.5 to < 3.0 x ULN	3.0 to < 5.0 x ULN	≥ 5.0 x ULN
Lipid Disorders (mg/dL; *mmol/L*)				
Cholesterol, Fasting, High				
≥ 18 years of age	200 to < 240 *5.18 to < 6.19*	240 to < 300 *6.19 to < 7.77*	≥ 300 *≥ 7.77*	NA
< 18 years of age	170 to < 200 *4.40 to < 5.15*	200 to < 300 *5.15 to < 7.77*	≥ 300 *≥ 7.77*	NA
LDL, Fasting, High				
≥ 18 years of age	130 to < 160 *3.37 to < 4.12*	160 to < 190 *4.12 to < 4.90*	≥ 190 *≥4.90*	NA
> 2 to < 18 years of age	110 to < 130 *2.85 to < 3.34*	130 to < 190 *3.34 to < 4.90*	≥ 190 *≥ 4.90*	NA
Triglycerides, Fasting, High	150 to 300 *1.71 to 3.42*	>300 to 500 *>3.42 to 5.7*	>500 to < 1,000 *>5.7 to 11.4*	> 1,000 *> 11.4*
Magnesium[15], Low (mEq/L; *mmol/L*)	1.2 to < 1.4 *0.60 to < 0.70*	0.9 to < 1.2 *0.45 to < 0.60*	0.6 to < 0.9 *0.30 to < 0.45*	< 0.6 *< 0.30*
Phosphate, Low (mg/dL; *mmol/L*)				
> 14 years of age	2.0 to < LLN *0.65 to < LLN*	1.4 to < 2.0 *0.45 to < 0.65*	1.0 to < 1.4 *0.32 to < 0.45*	< 1.0 *< 0.32*
1 to 14 years of age	3.0 to < 3.5 *0.97 to < 1.13*	2.5 to < 3.0 *0.81 to < 0.97*	1.5 to < 2.5 *0.48 to < 0.81*	< 1.5 *< 0.48*
< 1 year of age	3.5 to < 4.5 *1.13 to < 1.45*	2.5 to < 3.5 *0.81 to < 1.13*	1.5 to < 2.5 *0.48 to < 0.81*	< 1.5 *< 0.48*
Potassium, High (mEq/L; *mmol/L*)	5.6 to < 6.0 *5.6 to < 6.0*	6.0 to < 6.5 *6.0 to < 6.5*	6.5 to < 7.0 *6.5 to < 7.0*	≥ 7.0 *≥ 7.0*
Potassium, Low (mEq/L; *mmol/L*)	3.0 to < 3.4 *3.0 to < 3.4*	2.5 to < 3.0 *2.5 to < 3.0*	2.0 to < 2.5 *2.0 to < 2.5*	< 2.0 *< 2.0*

[15] To convert a magnesium value from mg/dL to mmol/L, laboratories should multiply by 0.4114.

Chemistries

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Sodium, High (mEq/L; *mmol/L*)	146 to < 150 *146 to < 150*	150 to < 154 *150 to < 154*	154 to < 160 *154 to < 160*	≥ 160 *≥ 160*
Sodium, Low (mEq/L; *mmol/L*)	130 to < 135 *130 to < 135*	125 to < 130 *125 to < 130*	121 to < 125 *121 to < 125*	≤ 120 *≤ 120*
Uric Acid, High (mg/dL; *mmol/L*)	7.5 to < 10.0 *0.45 to < 0.59*	10.0 to < 12.0 *0.59 to < 0.71*	12.0 to < 15.0 *0.71 to < 0.89*	≥ 15.0 *≥ 0.89*

Hematology

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Absolute CD4+ Count, Low (cell/mm^3; *cells/L*)				
> 5 years of age (not HIV infected)	300 to < 400 *300 to < 400*	200 to < 300 *200 to < 300*	100 to < 200 *100 to < 200*	< 100 *< 100*
Absolute Lymphocyte Count, Low (cell/mm^3; *cells/L*)				
> 5 years of age (not HIV infected)	600 to < 650 *0.600 x 10^9 to < 0.650 x 10^9*	500 to < 600 *0.500 x 10^9 to < 0.600 x 10^9*	350 to < 500 *0.350 x 10^9 to < 0.500 x 10^9*	< 350 *< 0.350 x 10^9*
Absolute Neutrophil Count (ANC), Low (cells/mm^3; *cells/L*)				
> 7 days of age	800 to 1,000 *0.800 x 10^9 to 1.000 x 10^9*	600 to 799 *0.600 x 10^9 to 0.799 x 10^9*	400 to 599 *0.400 x 10^9 to 0.599 x 10^9*	< 400 *< 0.400 x 10^9*
2 to 7 days of age	1,250 to 1,500 *1.250 x 10^9 to 1.500 x 10^9*	1,000 to 1,249 *1.000 x 10^9 to 1.249 x 10^9*	750 to 999 *0.750 x 10^9 to 0.999 x 10^9*	< 750 *< 0.750 x 10^9*
≤ 1 day of age	4,000 to 5,000 *4.000 x 10^9 to 5.000 x 10^9*	3,000 to 3,999 *3.000 x 10^9 to 3.999 x 10^9*	1,500 to 2,999 *1.500 x 10^9 to 2.999 x 10^9*	< 1,500 *< 1.500 x 10^9*
Fibrinogen, Decreased (mg/dL; *g/L*)	100 to < 200 *1.00 to < 2.00* OR 0.75 to < 1.00 x LLN	75 to < 100 *0.75 to < 1.00* OR ≥ 0.50 to < 0.75 x LLN	50 to < 75 *0.50 to < 0.75* OR 0.25 to < 0.50 x LLN	< 50 *< 0.50* OR < 0.25 x LLN OR Associated with gross bleeding
Hemoglobin[16], Low (g/dL; *mmol/L*) [17]				
≥ 13 years of age (male only)	10.0 to 10.9 *6.19 to 6.76*	9.0 to < 10.0 *5.57 to < 6.19*	7.0 to < 9.0 *4.34 to < 5.57*	< 7.0 *< 4.34*
≥ 13 years of age (female only)	9.5 to 10.4 *5.88 to 6.48*	8.5 to < 9.5 *5.25 to < 5.88*	6.5 to < 8.5 *4.03 to < 5.25*	< 6.5 *< 4.03*

[16] Male and female sex are defined as sex at birth. For transgender participants ≥13 years of age who have been on hormone therapy for more than 6 consecutive months, grade hemoglobin based on the gender with which they identify (i.e., a transgender female should be graded using the female sex at birth hemoglobin laboratory values).
[17] The most commonly used conversion factor to convert g/dL to mmol/L is 0.6206. For grading hemoglobin results obtained by an analytic method with a conversion factor other than 0.6206, the result must be converted to g/dL using appropriate conversion factor for the particular laboratory.

Hematology

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
57 days of age to < 13 years of age (male and female)	9.5 to 10.4 *5.88 to 6.48*	8.5 to < 9.5 *5.25 to < 5.88*	6.5 to < 8.5 *4.03 to < 5.25*	< 6.5 *< 4.03*
36 to 56 days of age (male and female)	8.5 to 9.6 *5.26 to 5.99*	7.0 to < 8.5 *4.32 to < 5.26*	6.0 to < 7.0 *3.72 to < 4.32*	< 6.0 *< 3.72*
22 to 35 days of age (male and female)	9.5 to 11.0 *5.88 to 6.86*	8.0 to < 9.5 *4.94 to < 5.88*	6.7 to < 8.0 *4.15 to < 4.94*	< 6.7 *< 4.15*
8 to ≤ 21 days of age (male and female)	11.0 to 13.0 *6.81 to 8.10*	9.0 to < 11.0 *5.57 to < 6.81*	8.0 to < 9.0 *4.96 to < 5.57*	< 8.0 *< 4.96*
≤ 7 days of age (male and female)	13.0 to 14.0 *8.05 to 8.72*	10.0 to < 13.0 *6.19 to < 8.05*	9.0 to < 10.0 *5.59 to < 6.19*	< 9.0 *< 5.59*
INR, High (not on anticoagulation therapy)	1.1 to < 1.5 x ULN	1.5 to < 2.0 x ULN	2.0 to < 3.0 x ULN	≥ 3.0 x ULN
Methemoglobin (% hemoglobin)	5.0 to < 10.0%	10.0 to < 15.0%	15.0 to < 20.0%	≥ 20.0%
PTT, High (not on anticoagulation therapy)	1.1 to < 1.66 x ULN	1.66 to < 2.33 x ULN	2.33 to < 3.00 x ULN	≥ 3.00 x ULN
Platelets, Decreased (cells/mm^3; *cells/L*)	100,000 to < 125,000 *100.000 x 10^9 to < 125.000 x 10^9*	50,000 to < 100,000 *50.000 x 10^9 to < 100.000 x 10^9*	25,000 to < 50,000 *25.000 x 10^9 to < 50.000 x 10^9*	< 25,000 *< 25.000 x 10^9*
PT, High (not on anticoagulation therapy	1.1 to < 1.25 x ULN	1.25 to < 1.50 x ULN	1.50 to < 3.00 x ULN	≥ 3.00 x ULN
WBC, Decreased (cells/mm^3; *cells/L*)				
> 7 days of age	2,000 to 2,499 *2.000 x 10^9 to 2.499 x 10^9*	1,500 to 1,999 *1.500 x 10^9 to 1.999 x 10^9*	1,000 to 1,499 *1.000 x 10^9 to 1.499 x 10^9*	< 1,000 *< 1.000 x 10^9*
≤ 7 days of age	5,500 to 6,999 *5.500 x 10^9 to 6.999 x 10^9*	4,000 to 5,499 *4.000 x 10^9 to 5.499 x 10^9*	2,500 to 3,999 *2.500 x 10^9 to 3.999 x 10^9*	< 2,500 *< 2.500 x 10^9*

Urinalysis

PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Glycosuria (random collection tested by dipstick)	Trace to 1+ or ≤ 250 mg	2+ or > 250 to ≤ 500 mg	> 2+ or > 500 mg	NA
Hematuria (not to be reported based on dipstick findings or on blood believed to be of menstrual origin)	6 to < 10 RBCs per high power field	≥ 10 RBCs per high power field	Gross, with or without clots OR With RBC casts OR Intervention indicated	Life-threatening consequences
Proteinuria (random collection tested by dipstick)	1+	2+	3+ or higher	NA

Appendix A
Total Bilirubin Table for Term and Preterm Neonates

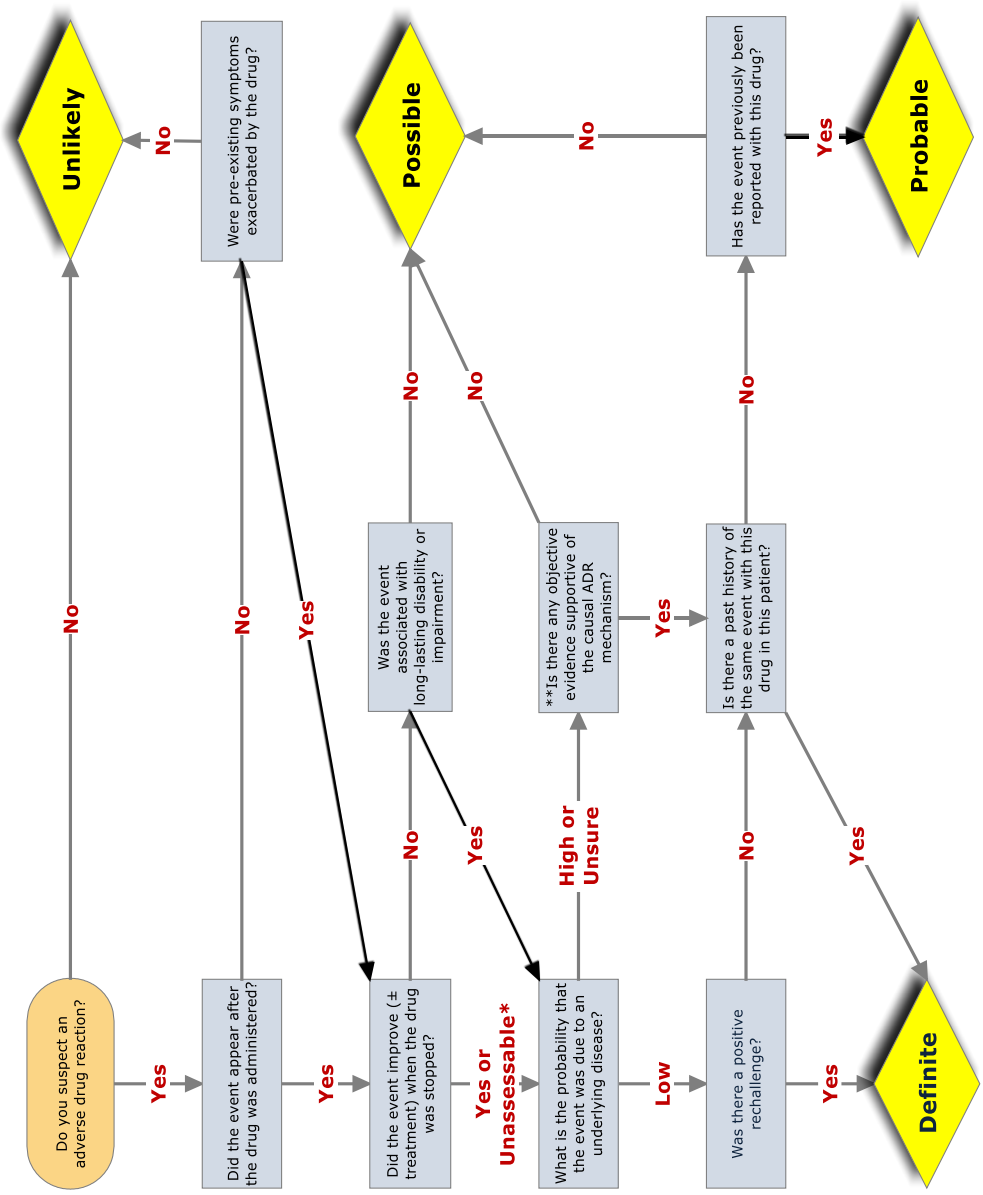
PARAMETER	GRADE 1 MILD	GRADE 2 MODERATE	GRADE 3 SEVERE	GRADE 4 POTENTIALLY LIFE-THREATENING
Total Bilirubin[18], High (mg/dL; *µmol/L*) [19]				
Term Neonate[20] *< 24 hours of age*	4 to < 7 *68.4 to < 119.7*	7 to < 10 *119.7 to < 171*	10 to < 17 *171* to < *290.7*	≥ 17 ≥ *290.7*
24 to < 48 hours of age	5 to < 8 *85.5 to < 136.8*	8 to < 12 *136.8 to < 205.2*	12 to < 19 *205.2* to < *324.9*	≥ 19 ≥ *324.9*
48 to < 72 hours of age	8.5 to < 13 *145.35 to < 222.3*	13 to < 15 *222.3 to < 256.5*	15 to < 22 *256.5* to < *376.2*	≥ 22 ≥ *376.2*
72 hours to < 7 days of age	11 to < 16 *188.1 to < 273.6*	16 to < 18 *273.6 to < 307.8*	18 to < 24 *307.8* to < *410.4*	≥ 24 ≥ *410.4*
7 to 28 days of age (breast feeding)	5 to < 10 *85.5 to < 171*	10 to < 20 *171 to < 342*	20 to < 25 *342 to < 427.5*	≥ 25 ≥ *427.5*
7 to 28 days of age (not breast feeding)	1.1 to < 1.6 x ULN	1.6 to < 2.6 x ULN	2.6 to < 5.0 x ULN	≥ 5.0 x ULN
Preterm Neonate[20] *35 to < 37 weeks gestational age*	Same as for ***Total Bilirubin, High, Term Neonate*** (based on days of age).	Same as for ***Total Bilirubin, High, Term Neonate*** (based on days of age).	Same as for ***Total Bilirubin, High, Term Neonate*** (based on days of age).	Same as for ***Total Bilirubin, High, Term Neonate*** (based on days of age).
32 to < 35 weeks gestational age and < 7 days of age	NA	NA	10 to < 14 *171 to < 239.4*	≥ 14 ≥ *239.4*
28 to < 32 weeks gestational age and < 7 days of age	NA	NA	6 to < 10 *102.6 to < 171*	≥ 10 ≥ *171*
< 28 weeks gestational age and < 7 days of age	NA	NA	5 to < 8 *85.5 to < 136.8*	≥ 8 ≥ *136.8*
7 to 28 days of age (breast feeding)	5 to < 10 *85.5 to < 171*	10 to < 20 *171 to < 342*	20 to < 25 *342 to < 427.5*	≥ 25 ≥ *427.5*
7 to 28 days of age (not breast feeding)	1.1 to < 1.6 x ULN	1.6 to < 2.6 x ULN	2.6 to < 5.0 x ULN	≥ 5.0 x ULN

[18] Severity grading for total bilirubin in neonates is complex because of rapidly changing total bilirubin normal ranges in the first week of life followed by the benign phenomenon of breast milk jaundice after the first week of life. Severity grading in this appendix corresponds approximately to cut-offs for indications for phototherapy at grade 3 and for exchange transfusion at grade 4.

[19] A laboratory value of 1 mg/dL is equivalent to 17.1 µmol/L.

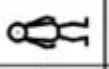
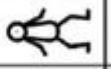
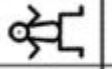
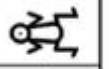
[20] Definitions: Term is defined as ≥ 37 weeks gestational age; near-term, as ≥ 35 weeks gestational age; preterm, as < 35 weeks gestational age; and neonate, as 0 to 28 days of age.

Appendix 6 Liverpool Causality Assessment Tool for Adverse Drug Reactions



Appendix 7 Ballard Score

Neuromuscular Maturity

Score	-1	0	1	2	3	4	5
Posture							
Square window (wrist)	 >90°	 90°	 60°	 45°	 30°	 0°	
Arm recoil		 180°	 140°–180°	 110°–140°	 90°–110°	 <90°	
Popliteal angle	 180°	 160°	 140°	 120°	 100°	 90°	 <90°
Scarf sign							
Heel to ear							

Physical Maturity

Skin	Sticky, friable, transparent	Gelatinous, red, translucent	Smooth, pink; visible veins	Superficial peeling and/or rash; few veins	Cracking, pale areas; rare veins	Parchment, deep cracking; no vessels	Leathery, cracked wrinkled
Lanugo	None	Sparse	Abundant	Thinning	Bald areas	Mostly bald	
Plantar surface	Heel-toe 40-50 mm: –1 <40 mm: –2	>50 mm, no crease	Faint red marks	Anterior transverse crease only	Creases anterior ⅔	Creases over entire sole	
Breast	Imperceptible	Barely perceptible	Flat areola, no bud	Stippled areola, 1–2 mm bud	Raised areola, 3–4 mm bud	Full areola, 5–10 mm bud	
Eye/Ear	Lids fused loosely: –1 tightly: –2	Lids open; pinna flat; stays folded	Slightly curved pinna; soft; slow recoil	Well curved pinna; soft but ready recoil	Formed and firm, instant recoil	Thick cartilage, ear stiff	
Genitals (male)	Scrotum flat, smooth	Scrotum empty, faint rugae	Testes in upper canal, rare rugae	Testes descending, few rugae	Testes down, good rugae	Testes pendulous, deep rugae	
Genitals (female)	Clitoris prominent, labia flat	Clitoris prominent, small labia minora	Clitoris prominent, enlarging minora	Majora and minora equally prominent	Majora large, minora small	Majora cover clitoris and minora	

Maturity Rating

Score	Weeks
-10	20
-5	22
0	24
5	26
10	28
15	30
20	32
25	34
30	36
35	38
40	40
45	42
50	44

Appendix 8 MOS-HIV Score

MOS-HIV 35-ITEM INSTRUMENT

INSTRUCTIONS TO PATIENT: Please answer the following questions by placing a "x" in the appropriate box.

1. In general, would you say that your health is:

(check one)

Excellent	1☐
Very Good	2☐
Good	3☐
Fair	4☐
Poor	5☐

2. How much **bodily** pain have you generally had during **the past 4 weeks**?

(check one)

None	1☐
Very Mild	2☐
Mild	3☐
Moderate	4☐
Severe	5☐
Very Severe	6☐

3. During **the past 4 weeks**, how much did **pain** interfere with your normal work (including both work outside the home and housework)?

(check one)

Not at all	1☐

A little bit 2☐

Moderately 3☐

Quite a bit 4☐

Extremely 5☐

4. The following questions are about activities you might do during a typical day. Does your health now limit you in these activities? If so, how much?

		(check one box on each line)		
		YES , Limited A Lot	**YES , Limited A Little**	**NO, Not Limited At All**
		1	**2**	**3**
a.	The kinds or amounts of **vigorous** activities you can do, like lifting heavy objects, running or participating in strenuous sports.	☐	☐	☐
b.	The kinds or amounts of **moderate** activities you can do, like moving a table, carrying groceries or bowling.	☐	☐	☐
c.	Walking uphill or climbing a few flights of stairs.	☐	☐	☐
d.	Bending, lifting or stooping.	☐	☐	☐
e.	Walking one block.	☐	☐	☐
f.	Eating, dressing, bathing, or using the toilet.	☐	☐	☐

5. Does your health **keep** you from working at a job, doing work around the house or going to school?

(check one)

Yes 1☐

No 2☐

6. Have you been unable to do **certain kinds or amounts** of work, housework, or schoolwork because of your health?

(check one)

Yes 1☐

No 2☐

For <u>each</u> of the following questions, please check the box for the <u>one</u> answer that comes <u>closest</u> to the way you have been feeling <u>during the past 4 weeks</u>.

(mark one box on each line)

	All of the Time	Most of the Time	A Good Bit of the Time	Some of the Time	A Little of the Time	None of the Time
	1	2	3	4	5	6
7. How much of the time, during the past 4 weeks, has your **health limited your social activities** (like visiting with friends or close relatives)?	☐	☐	☐	☐	☐	☐
8. How much of the time, during the past 4 weeks:						
a. Have you been a very nervous person?	☐	☐	☐	☐	☐	☐
b. Have you felt calm and peaceful?	☐	☐	☐	☐	☐	☐
c. Have you felt downhearted and blue?	☐	☐	☐	☐	☐	☐
d. Have you been a happy person?	☐	☐	☐	☐	☐	☐
e. Have you felt so down in the dumps that nothing could cheer you up?	☐	☐	☐	☐	☐	☐

(check one box on each line)

	All Of the Time	Most of the time	A Good Bit of the Time	Some of the Time	A Little of the Time	None of the Time
	1	2	3	4	5	6
9. How often during the **past four weeks**:						
a. Did you feel full of pep?	☐	☐	☐	☐	☐	☐
b. Did you feel worn out?	☐	☐	☐	☐	☐	☐
c. Did you feel tired?	☐	☐	☐	☐	☐	☐
d. Did you have enough energy to do the things you wanted to do?	☐	☐	☐	☐	☐	☐
e. Did you feel weighed down by your health problems?	☐	☐	☐	☐	☐	☐
f. Were you discouraged by your health problems?	☐	☐	☐	☐	☐	☐
g. Did you feel despair over your health problems?	☐	☐	☐	☐	☐	☐
h. Were you afraid because of your health?	☐	☐	☐	☐	☐	☐

(check one box on each line)

	All of the Time	Most of the Time	A Good Bit of the Time	Some of the Time	A Little of the Time	None of the Time
	1	2	3	4	5	6
10. How much of the time, during the **past 4 weeks**:						
a. Did you have difficulty reasoning and solving problems, for example, making plans, making decisions, learning new things?	☐	☐	☐	☐	☐	☐
b. Did you forget things that happened recently, for example, where you put things and when you had appointments?	☐	☐	☐	☐	☐	☐
c. Did you have trouble keeping your attention on any activity for long?	☐	☐	☐	☐	☐	☐
d. Did you have difficulty doing activities involving concentration and thinking?	☐	☐	☐	☐	☐	☐

(check one box on each line)

	Definitely True	Mostly True	Don't Know	Mostly False	Definitely False
	1	2	3	4	5
11. Please check the box that describes whether each of the following statements is true or false for you.					
a. I am somewhat ill.	☐	☐	☐	☐	☐
b. I am as healthy as anybody I know.	☐	☐	☐	☐	☐

c. My health is excellent.	☐	☐	☐	☐	☐
d. I have been feeling bad lately.	☐	☐	☐	☐	☐

12. How has the quality of your life been during the **past 4 weeks**? That is, how have things been going for you?

(check one)

Very well; could hardly be better	1☐
Pretty good	2☐
Good and bad parts about equal	3☐
Pretty bad	4☐
Very bad; could hardly be worse	5☐

13. How would you rate your physical health and emotional condition now compared to **4 weeks ago**?

(check one)

Much better	1☐
A little better	2☐
About the same	3☐
A little worse	4☐
Much worse	5☐

Appendix 9 EQ-5D-5L

Under each heading, please tick the ONE box that best describes your health TODAY.

MOBILITY

I have no problems in walking about ❑
I have slight problems in walking about ❑
I have moderate problems in walking about ❑
I have severe problems in walking about ❑
I am unable to walk about ❑

SELF-CARE

I have no problems washing or dressing myself ❑
I have slight problems washing or dressing myself ❑
I have moderate problems washing or dressing myself ❑
I have severe problems washing or dressing myself ❑
I am unable to wash or dress myself ❑

USUAL ACTIVITIES *(e.g. work, study, housework, family or leisure activities)*

I have no problems doing my usual activities ❑
I have slight problems doing my usual activities ❑
I have moderate problems doing my usual activities ❑
I have severe problems doing my usual activities ❑
I am unable to do my usual activities ❑

PAIN / DISCOMFORT

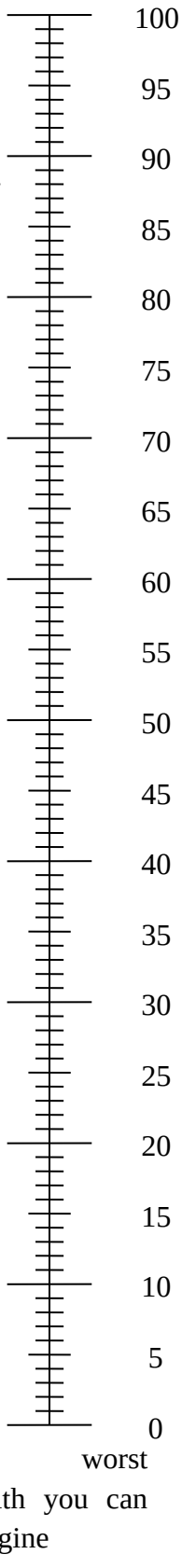
I have no pain or discomfort ❑
I have slight pain or discomfort ❑
I have moderate pain or discomfort ❑
I have severe pain or discomfort ❑
I have extreme pain or discomfort ❑

ANXIETY / DEPRESSION

I am not anxious or depressed ❑
I am slightly anxious or depressed ❑
I am moderately anxious or depressed ❑
I am severely anxious or depressed ❑
I am extremely anxious or depressed ❑

We would like to know how good or bad your health is TODAY.
This scale is numbered from 0 to 100.
100 means the best health you can imagine.
0 means the worst health you can imagine.
Mark an X on the scale to indicate how your health is TODAY.
Now, please write the number you marked on the scale in the box below.

YOUR HEALTH TODAY = ☐



Appendix 10 Pittsburgh Sleep Quality Index

PITTSBURGH SLEEP QUALITY INDEX
INSTRUCTIONS:
The following questions relate to your usual sleep habits during the past month only. Your answers should indicate the most accurate reply for the majority of days and nights in the past month.
Please answer all questions.
1. During the past month, what time have you usually gone to bed at night?

BED TIME _____

2. During the past month, how long (in minutes) has it usually taken you to fall asleep each night?

NUMBER OF MINUTES _____

3. During the past month, what time have you usually gotten up in the morning?

GETTING UP TIME _____

4. During the past month, how many hours of actual sleep did you get at night? (This may be different than the number of hours you spent in bed.)

HOURS OF SLEEP PER NIGHT _____

For each of the remaining questions, check the one best response. Please answer all questions.

5. During the past month, how often have you had trouble sleeping because you . . .

a) Cannot get to sleep within 30 minutes

Not during the past month	Less than once a week	Once or twice a week	Three or more times a week
_____	_____	_____	_____

b) Wake up in the middle of the night or early morning

Not during the past month	Less than once a week	Once or twice a week	Three or more times a week
_____	_____	_____	_____

c) Have to get up to use the bathroom

Not during the past month	Less than once a week	Once or twice a week	Three or more times a week
_____	_____	_____	_____

d) Cannot breathe comfortably

Not during the past month	Less than once a week	Once or twice a week	Three or more times a week
_____	_____	_____	_____

e) Cough or snore loudly

Not during the past month	Less than once a week	Once or twice a week	Three or more times a week
_____	_____	_____	_____

f) Feel too cold

Not during the past month	Less than once a week	Once or twice a week	Three or more times a week
_____	_____	_____	_____

g) Feel too hot

Not during the	Less than	Once or twice	Three or more

Buysse DJ, Reynolds CF, Monk TH, Berman SR, Kupfer DJ: Psychiatry Research, 28:193-213, 1989.

past month_____ once a week_____ a week_____ times a week_____
h) Had bad dreams
Not during the Less than Once or twice Three or more
past month_____ once a week_____ a week_____ times a week_____
i) Have pain
Not during the Less than Once or twice Three or more
past month_____ once a week_____ a week_____ times a week_____
j) Other reason(s), please describe_____
_____
How often during the past month have you had trouble sleeping because of this?
Not during the Less than Once or twice Three or more
past month_____ once a week_____ a week_____ times a week_____
6. During the past month, how would you rate your sleep quality overall?
Very good _____
Fairly good _____
Fairly bad _____
Very bad _____
7. During the past month, how often have you taken medicine to help you sleep (prescribed or "over the counter")?
Not during the Less than Once or twice Three or more
past month_____ once a week_____ a week_____ times a week_____
8. During the past month, how often have you had trouble staying awake while driving, eating meals, or engaging in social activity?
Not during the Less than Once or twice Three or more
past month_____ once a week_____ a week_____ times a week_____
9. During the past month, how much of a problem has it been for you to keep up enough enthusiasm to get things done?
No problem at all _____
Only a very slight problem _____
Somewhat of a problem _____
A very big problem _____
10. Do you have a bed partner or room mate?
No bed partner or room mate _____
Partner/room mate in other room _____
Partner in same room, but not same bed _____
Partner in same bed _____
If you have a room mate or bed partner, ask him/her how often in the past month you have had . . .
a) Loud snoring
Not during the Less than Once or twice Three or more
past month_____ once a week_____ a week_____ times a week_____

Buysse DJ, Reynolds CF, Monk TH, Berman SR, Kupfer DJ: Psychiatry Research, 28:193-213, 1989.

b) Long pauses between breaths while asleep

Not during the past month_____ Less than once a week_____ Once or twice a week_____ Three or more times a week_____

c) Legs twitching or jerking while you sleep

Not during the past month_____ Less than once a week_____ Once or twice a week_____ Three or more times a week_____

d) Episodes of disorientation or confusion during sleep

Not during the past month_____ Less than once a week_____ Once or twice a week_____ Three or more times a week_____

e) Other restlessness while you sleep; please describe_____

_____

Not during the past month_____ Less than once a week_____ Once or twice a week_____ Three or more times a week_____

Buysse DJ, Reynolds CF, Monk TH, Berman SR, Kupfer DJ: Psychiatry Research, 28:193-213, 1989.

Appendix 11 Social Economic Questionnaire

1. Record the time of interview:	hr min 24-hr clock
2. How long have you been living continuously in the city /town or village your currently living in? (***If less than one year, record "00" years.)***	
3. Name of current city, town or village of residence:	
4. Do you always live here or as a visitor?	Always Visitor
5. Just before you moved here, did you live in a city, in a town, or in a rural area?	City Town Rural Area
6. Before you moved here, which ***(Province/Region/State)*** did you live in?	
7. What is your ethnicity?	
8. How old were you at your last birthday?	***Age in completed years***
9. Have you ever attended school?	Yes No
10. If ***'yes'*** what is the highest level of school you attended:	Primary Secondary Higher Tertiary
11. What is the highest ***(Grade/Form/Year)*** you completed at that level? ***(If completed less than one year at that level, record "00")***	***Grade/Form/Year***

12. Are you currently employed:	Yes No
13. Are you married?	Yes No
13a. If '***yes'*** are you currently living with your partner?	Yes No
13b. Since the last study visit has anything bad happened to you as a result of taking part in the trial?	Yes No
14. Do you read a newspaper or magazine at least once a week, less than once a week or not at all?	At least once a week Less than once a week Not at all
15. Do you listen to the radio at least once a week, less than once a week or not at all?	At least once a week Less than once a week Not at all
16. Do you watch television at least once a week, less than once a week or not at all?	At least once a week Less than once a week Not at all
17. Do you own a mobile telephone?	Yes No
18. Do you use a mobile phone for any financial transactions?	Yes No
19. Do you have an account in a bank or other financial institution that you yourself use?	Yes No
20. Have you ever used the internet?	Yes No

21. In the last 12 months, have you used the internet?	Yes No
22. During the last one month, how often did you use the internet:	Almost every day At least once a week Less than once a week Not at all
23. In the last 12 months, how many times have you been away from home for one or more nights?	***Number of times*** None
24. In the last 12 months, have you been away from home for more than one months at a times?	Yes No

Appendix 12 CBCL Questionnaire

Please print. **CHILD BEHAVIOR CHECKLIST FOR AGES 1½-5**

For office use only
ID #

CHILD'S FULL NAME First Middle Last

CHILD'S GENDER ☐ Boy ☐ Girl

CHILD'S AGE

CHILD'S ETHNIC GROUP OR RACE

TODAY'S DATE Mo._____ Day_____ Year_____

CHILD'S BIRTHDATE Mo._____ Day_____ Year_____

Please fill out this form to reflect your view of the child's behavior even if other people might not agree. Feel free to write additional comments beside each item and in the space provided on page 2. **Be sure to answer all items.**

PARENTS' USUAL TYPE OF WORK, even if not working now. Please be specific — for example, auto mechanic, high school teacher, homemaker, laborer, lathe operator, shoe salesman, army sergeant.

PARENT 1 (or MOTHER) TYPE OF WORK _____

PARENT 2 (or FATHER) TYPE OF WORK _____

THIS FORM FILLED OUT BY: (print your full name)

_____

Your relation to child:
☐ Parent 1 (or Mother) ☐ Parent 2 (or Father) ☐ Other (specify):

Below is a list of items that describe children. For each item that describes the child **now or within the past 2 months**, please circle the **2** if the item is **very true or often true** of the child. Circle the **1** if the item is **somewhat or sometimes true** of the child. If the item is **not true** of the child, circle the **0**. Please answer all items as well as you can, even if some do not seem to apply to the child.

0 = Not True (as far as you know) **1 = Somewhat or Sometimes True** **2 = Very True or Often True**

0 1 2 1. Aches or pains (without medical cause; **do not** include stomach or headaches)
0 1 2 2. Acts too young for age
0 1 2 3. Afraid to try new things
0 1 2 4. Avoids looking others in the eye
0 1 2 5. Can't concentrate, can't pay attention for long
0 1 2 6. Can't sit still, restless, or hyperactive
0 1 2 7. Can't stand having things out of place
0 1 2 8. Can't stand waiting; wants everything now
0 1 2 9. Chews on things that aren't edible
0 1 2 10. Clings to adults or too dependent
0 1 2 11. Constantly seeks help
0 1 2 12. Constipated, doesn't move bowels (when not sick)
0 1 2 13. Cries a lot
0 1 2 14. Cruel to animals
0 1 2 15. Defiant
0 1 2 16. Demands must be met immediately
0 1 2 17. Destroys his/her own things
0 1 2 18. Destroys things belonging to his/her family or other children
0 1 2 19. Diarrhea or loose bowels (when not sick)
0 1 2 20. Disobedient
0 1 2 21. Disturbed by any change in routine
0 1 2 22. Doesn't want to sleep alone
0 1 2 23. Doesn't answer when people talk to him/her
0 1 2 24. Doesn't eat well (describe): _____
0 1 2 25. Doesn't get along with other children
0 1 2 26. Doesn't know how to have fun; acts like a little adult
0 1 2 27. Doesn't seem to feel guilty after misbehaving
0 1 2 28. Doesn't want to go out of home
0 1 2 29. Easily frustrated
0 1 2 30. Easily jealous
0 1 2 31. Eats or drinks things that are not food—**don't** include sweets (describe): _____
0 1 2 32. Fears certain animals, situations, or places (describe): _____
0 1 2 33. Feelings are easily hurt
0 1 2 34. Gets hurt a lot, accident-prone
0 1 2 35. Gets in many fights
0 1 2 36. Gets into everything
0 1 2 37. Gets too upset when separated from parents
0 1 2 38. Has trouble getting to sleep
0 1 2 39. Headaches (without medical cause)
0 1 2 40. Hits others
0 1 2 41. Holds his/her breath
0 1 2 42. Hurts animals or people without meaning to
0 1 2 43. Looks unhappy without good reason
0 1 2 44. Angry moods
0 1 2 45. Nausea, feels sick (without medical cause)
0 1 2 46. Nervous movements or twitching (describe): _____
0 1 2 47. Nervous, highstrung, or tense
0 1 2 48. Nightmares
0 1 2 49. Overeating
0 1 2 50. Overtired
0 1 2 51. Shows panic for no good reason
0 1 2 52. Painful bowel movements (without medical cause)
0 1 2 53. Physically attacks people
0 1 2 54. Picks nose, skin, or other parts of body (describe): _____

Be sure you answered all items. Then see other side.

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Please print your answers. Be sure to answer all items.

0 = Not True (as far as you know) 1 = Somewhat or Sometimes True 2 = Very True or Often True

0 1 2 55. Plays with own sex parts too much
0 1 2 56. Poorly coordinated or clumsy
0 1 2 57. Problems with eyes (without medical cause) (describe): _____
0 1 2 58. Punishment doesn't change his/her behavior
0 1 2 59. Quickly shifts from one activity to another
0 1 2 60. Rashes or other skin problems (without medical cause)
0 1 2 61. Refuses to eat
0 1 2 62. Refuses to play active games
0 1 2 63. Repeatedly rocks head or body
0 1 2 64. Resists going to bed at night
0 1 2 65. Resists toilet training (describe): _____
0 1 2 66. Screams a lot
0 1 2 67. Seems unresponsive to affection
0 1 2 68. Self-conscious or easily embarrassed
0 1 2 69. Selfish or won't share
0 1 2 70. Shows little affection toward people
0 1 2 71. Shows little interest in things around him/her
0 1 2 72. Shows too little fear of getting hurt
0 1 2 73. Too shy or timid
0 1 2 74. Sleeps less than most kids during day and/or night (describe): _____
0 1 2 75. Smears or plays with bowel movements
0 1 2 76. Speech problem (describe): _____
0 1 2 77. Stares into space or seems preoccupied
0 1 2 78. Stomachaches or cramps (without medical cause)
0 1 2 79. Rapid shifts between sadness and excitement
0 1 2 80. Strange behavior (describe): _____
0 1 2 81. Stubborn, sullen, or irritable
0 1 2 82. Sudden changes in mood or feelings
0 1 2 83. Sulks a lot
0 1 2 84. Talks or cries out in sleep
0 1 2 85. Temper tantrums or hot temper
0 1 2 86. Too concerned with neatness or cleanliness
0 1 2 87. Too fearful or anxious
0 1 2 88. Uncooperative
0 1 2 89. Underactive, slow moving, or lacks energy
0 1 2 90. Unhappy, sad, or depressed
0 1 2 91. Unusually loud
0 1 2 92. Upset by new people or situations (describe): _____
0 1 2 93. Vomiting, throwing up (without medical cause)
0 1 2 94. Wakes up often at night
0 1 2 95. Wanders away
0 1 2 96. Wants a lot of attention
0 1 2 97. Whining
0 1 2 98. Withdrawn, doesn't get involved with others
0 1 2 99. Worries
0 1 2 100. Please write in any problems the child has that were not listed above.
0 1 2 _____
0 1 2 _____
0 1 2 _____

Please be sure you have answered all items. Underline any you are concerned about.

Does the child have any illness or disability (either physical or mental)? ☐ **No** ☐ **Yes—Please describe:**

What concerns you most about the child?

Please describe the best things about the child:

LANGUAGE DEVELOPMENT SURVEY FOR AGES 18-35 MONTHS

For office use only
ID #

The Language Development Survey assesses children's word combinations and vocabulary. By carefully completing the Language Development Survey, you can help us obtain an accurate picture of the child's developing language. ***Please print your answers. Be sure to answer all items.***

I Was the child born earlier than the usual 9 months after conception?

G No G YesChow many weeks early?_____weeks early.

II How much did the child weigh at birth?_____pounds_____ounces; or_____grams.

III How many ear infections did the child have before age 24 months?

G 0-2 G 3-5 G 6-8 G 9 or more

IV. Is any language beside English spoken in the child's home?

G No G Yes—please list the languages: _____ _____

_____ _____.

V. Has anyone in the child's family been slow in learning to talk?

G No G Yes—please list their relationships to the child; for example, brother, father:

_____

VI Are you worried about the child's language development?

G No G Yes—why? _____

_____

VII Does the child spontaneously say words in any language? (not just imitates or understands words)?

G No G Yes—if yes, please complete item VIII and page 4.

VIII Does the child combine 2 or more words into phrases? For example: "more cookie," "car bye-bye."

G No G Yes—please print 5 of the child-s longest and best phrases or sentences.

For each phrase that is not in English, print the name of the language.

1. _____
2. _____
3. _____
4. _____
5. _____

Be sure you answered all items. Then see other side.

PAGE 3

Please circle each word that the child says SPONTANEOUSLY (not just imitates or understands). If your child says non-English versions of words on the list, circle the English word and write the first letter of the language (e.g., S for Spanish). Please include words even if they are not pronounced clearly or are in "baby talk" (for example: "baba" for bottle).

FOODS

1. apple
2. banana
3. bread
4. butter
5. cake
6. candy
7. cereal
8. cheese
9. coffee
10. cookie
11. crackers
12. drink
13. egg
14. food
15. grapes
16. gum
17. hamburger
18. hotdog
19. ice cream
20. juice
21. meat
22. milk
23. orange
24. pizza
25. pretzel
26. raisins
27. soda
28. soup
29. spaghetti
30. tea
31. toast
32. water

TOYS

33. ball
34. balloon
35. blocks
36. book
37. crayons
38. doll
39. picture
40. present
41. slide
42. swing
43. teddy bear

OUTDOORS

44. flower
45. house
46. moon
47. rain
48. sidewalk
49. sky
50. snow
51. star
52. street
53. sun
54. tree

ANIMALS

55. bear
56. bee
57. bird
58. bug
59. bunny
60. cat
61. chicken
62. cow
63. dog
64. duck
65. elephant
66. fish
67. frog
68. horse
69. monkey
70. pig
71. puppy
72. snake
73. tiger
74. turkey
75. turtle

BODY PARTS

76. arm
77. belly button
78. bottom
79. chin
80. ear
81. elbow
82. eye
83. face
84. finger
85. foot
86. hair
87. hand
88. knee
89. leg
90. mouth
91. neck
92. nose
93. teeth
94. thumb
95. toe
96. tummy

VEHICLES

97. bike
98. boat
99. bus
100. car
101. motorcycle
102. plane
103. stroller
104. train
105. trolley
106. truck

ACTIONS

107. bath
108. breakfast
109. bring
110. catch
111. clap
112. close
113. come
114. cough
115. cut
116. dance
117. dinner
118. doodoo/poop
119. down
120. eat
121. feed
122. finish
123. fix
124. get
125. give
126. go
127. have
128. help
129. hit
130. hug
131. jump
132. kick
133. kiss
134. knock
135. look
136. love
137. lunch
138. make
139. nap
140. open
141. outside
142. pattycake
143. peekaboo
144. peepee
145. push
146. read
147. ride
148. run
149. see
150. show
151. shut
152. sing
153. sit
154. sleep
155. stop
156. take
157. throw
158. tickle
159. up
160. walk
161. want
162. wash

HOUSEHOLD

163. bathtub
164. bed
165. blanket
166. bottle
167. bowl
168. chair
169. clock
170. crib
171. cup
172. door
173. floor
174. fork
175. glass
176. knife
177. light
178. mirror
179. pillow
180. plate
181. potty
182. radio
183. room
184. sink
185. soap
186. spoon
187. stairs
188. table
189. telephone
190. towel
191. trash
192. T.V.
193. window

PERSONAL

194. brush
195. comb
196. glasses
197. key
198. money
199. paper
200. pen
201. pencil
202. penny
203. pocketbook
204. tissue
205. toothbrush
206. umbrella
207. watch

PLACES

208. church
209. home
210. hospital
211. library
212. park
213. school
214. store
215. zoo

MODIFIERS

216. all gone
217. all right
218. bad
219. big
220. black
221. blue
222. broken
223. clean
224. cold
225. dark
226. dirty
227. dry
228. good
229. happy
230. heavy
231. hot
232. hungry
233. little
234. mine
235. more
236. nice
237. pretty
238. red
239. stinky
240. that
241. this
242. tired
243. wet
244. white
245. yellow
246. yucky

CLOTHES

247. belt
248. boots
249. coat
250. diaper
251. dress
252. gloves
253. hat
254. jacket
255. mittens
256. pajamas
257. pants
258. shirt
259. shoes
260. slippers
261. sneakers
262. socks
263. sweater

OTHER

264. any letter
265. away
266. booboo
267. byebye
268. excuse me
269. here
270. hi, hello
271. in
272. me
273. meow
274. my
275. myself
276. nightnight
277. no
278. off
279. on
280. out
281. please
282. Sesame St.
283. shut up
284. thank you
285. there
286. under
287. welcome
288. what
289. where
290. why
291. woofwoof
292. yes
293. you
294. yumyum
295. any number

PEOPLE

296. aunt
297. baby
298. boy
299. daddy
300. doctor
301. girl
302. grandma
303. grandpa
304. lady
305. man
306. mommy
307. own name
308. pet name
309. uncle
310. name of TV or story character

Other words your child says, including non-English words:

_____

_____

_____